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

A. 510(k) Number:
k131168

B. Purpose for Submission:
New device

C. Measurand:
Oxycodone

D. Type of Test:
Homogenous enzyme immunoassay

E. Applicant:
Immunalysis Corporation

F. Proprietary and Established Names:
Immunalysis Oxycodone Urine Enzyme Immunoassay
Immunalysis Oxycodone Urine Calibrators
Immunalysis Oxycodone Urine Controls

G. Regulatory Information:

1. Regulation section:
   21 CFR § 862.3650 Opiate Test System
   21 CFR § 862.3200 Clinical Toxicology Calibrator
   21 CFR § 862.3280 Clinical Toxicology Control Material

2. Classification:
   Class II for 862.3650 and 862.3200
   Class I, reserved for 862.3280

3. Product code:
   DJG Enzyme Immunoassay, Opiates
   DLJ Calibrators, Drug Specific
   LAS Drug Specific Control Materials

4. Panel:
   Toxicology (91)

H. Intended Use:

1. Intended use(s):
   The Immunalysis Oxycodone Urine Enzyme Immunoassay is a homogenous enzyme immunoassay with a dual cutoff of 100 ng/mL and 300 ng/mL. The

assay is intended for use in laboratories for the qualitative and semi-quantitative analysis of Oxycodone in human urine with automated clinical chemistry analyzers. This assay is calibrated against Oxycodone. This in-vitro diagnostic device is for prescription use only. The semi-quantitative mode is for purposes of enabling laboratories to determine an appropriate dilution of the specimen for confirmation by a confirmatory method such as GC-MS or permitting laboratories to establish quality control procedures.

The Immunalysis Oxycodone Urine Enzyme Immunoassay Kit provides only a preliminary analytical test result. A more specific alternate chemical method must be used in order to obtain a confirmed analytical result. Gas Chromatography/Mass Spectrometry (GC-MS) or Liquid Chromatography/Mass Spectroscopy (LC/MS) is the preferred confirmatory method. Clinical consideration and professional judgment should be applied to any drug of abuse test result, particularly when preliminary positive results are used.

The Immunalysis Oxycodone Urine Calibrators are used as calibrators in the Immunalysis Oxycodone Urine Enzyme Immunoassay for the qualitative and semi-quantitative determination of Oxycodone in urine on automated clinical chemistry analyzers.

The Immunalysis Oxycodone Urine Controls are used as control materials in the Immunalysis Oxycodone Urine Enzyme Immunoassay.

2. Indication(s) for use:
   See intended uses above.

3. Special conditions for use statement(s):
   - For prescription use only
   - For *in vitro* diagnostic use only

4. Special instrument requirements:
   The Beckman Coulter AU400e Chemistry Analyzer was used to generate the performance data in this submission.

I. Device Description:

The assay consists of antibody/substrate reagent and enzyme conjugate reagent. The antibody/substrate reagent includes recombinant monoclonal antibodies to Oxycodone, glucose-6-phosphate (G6P) and nicotinamide adenine dinucleotide (NAD) in Tris buffer with Sodium Azide as a preservative. The enzyme conjugate reagent includes oxycodone derivative labeled with glucose-6-phosphate dehydrogenase (G6PDH) in Tris buffer with Sodium Azide as a preservative.

Calibrators and control materials are prepared from synthetic negative urine and a commercially available oxycodone drug standard. The concentration of the Oxycodone in these calibrators and control materials is determined by Liquid

chromatography/Mass Spectroscopy (LC/MS).

The following calibrators are available:

- Oxycodone Calibrator Set (0, 100ng/mL, 300 ng/mL, 500ng/mL and 1000ng/mL) for the semi-quantitative mode.
- 100 ng/mL Oxycodone Calibrator for the 100ng/mL cutoff
- 300 ng/mL Oxycodone Calibrator for the 300ng/mL cutoff

The following control materials are available:

- Oxycodone Controls set for the 100ng/mL cutoff (75ng/mL and 125ng/mL)
- Oxycodone Control set for the 300ng/mL cutoff (225ng/mL and 375ng/mL)

Oxycodone Calibrators and controls are sold separately. Reagents are liquid, ready to use.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   DRI Oxycodone Assay, DRI Oxycodone Calibrators and DRI Oxycodone Controls

2. Predicate K number(s):
   k040411

3. Comparison with predicate:

Item	Immunalysis Oxycodone Urine Enzyme Immunoassay	Predicate k040411
Similarities		
Intended Use	For the qualitative and semi-quantitative analysis of Oxycodone in human urine at cutoffs of 100 and 300 ng/mL	same
Measured Analyte	Oxycodone	same
Test Matrix	Human urine	same
Cutoff Level	100 ng/mL and 300 ng/mL	same
Technology	Homogenous enzyme immunoassay	same
Differences		
Antibodies	Recombinant FAB antibody to oxycodone	Mouse monoclonal anti-oxycodone derivative

Item	Immunalysis Oxycodone Urine Calibrators	Predicate k040411
Similarities		
Intended Use	For the calibration of the the Immunalysis Oxycodone Urine	For the calibration of the DRI

Item	Immunalysis Oxycodone Urine Calibrators	Predicate k040411
	Enzyme Immunoassay	Oxycodone Assay
Calibrator Form	Liquid	same
Calibrator Levels	Five levels (0, 100, 300, 500 and 1000 ng/mL)	same

Item	Immunalysis Oxycodone Urine Controls	Predicate k040411
Similarities		
Intended Use	Intended as control materials for the Immunalysis Oxycodone Urine Enzyme Immunoassay	Intended as control materials for the DRI Oxycodone Assay
Control Levels	2 Levels (75 and 125 ng/mL or 225 and 375 ng/mL)	same

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

- Evaluation of Precision Performance of Quantitative Measurement Methods: Approved Guideline (EP5-A2)
- Interference Testing in Clinical Chemistry; Approved Guideline (EP7-A2)
- Draft Guidance for Industry and FDA Staff Premarket Submission and Labeling Recommendations for Drugs of Abuse Screening Tests

L. Test Principle:

The assay uses an Oxycodone specific antibody. The assay is based on the competition of Oxycodone labeled enzyme glucose-6-phosphate dehydrogenase (G6PDH) and the free drug in the urine sample for the fixed amount of antibody binding sites. In the absence of the free drug in the sample, the antibody binds the drug enzyme conjugate and enzyme activity is inhibited. This creates a dose response relationship between drug concentration in the urine sample and enzyme activity. The enzyme G6PDH activity is determined at 340 nm spectrophotometrically by the conversion of NAD to NADH.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:
   a. Precision/Reproducibility:
   The sponsor performed precision studies in-house following the guidelines provided in CLSI EP5-A2. The study was performed using drug free urine spiked with Oxycodone and 1 reagent lot using 1 Beckman Coulter AU400e Chemistry Analyzer. Samples were measured in duplicate on 2 runs per day for 20 days (n=80). The data are summarized in the following table:

Qualitative analysis (100ng/mL cutoff)

Concentration	% of cutoff	Result
0	-100%	80 negative
25	-75%	80 negative
50	-50%	80 negative
75	-25%	80 negative
100	Cutoff	46 negative 34 positive
125	+25%	80 positive
150	+50%	80 positive
175	+75%	80 positive
200	+100%	80 positive

Qualitative analysis (300ng/mL cutoff)

Concentration	% of cutoff	Result
0	-100%	80 negative
75	-75%	80 negative
150	-50%	80 negative
225	-25%	80 negative
300	Cutoff	35 negative 45 positive
375	+25%	80 positive
450	+50%	80 positive
525	+75%	80 positive
600	+100%	80 positive

Semi-Quantitative analysis (100ng/mL cutoff)

Concentration	% of cutoff	Result
0	-100%	80 negative
25	-75%	80 negative
50	-50%	80 negative
75	-25%	80 negative
100	Cutoff	57 negative 23 positive
125	+25%	80 positive
150	+50%	80 positive
175	+75%	80 positive
200	+100%	80 positive

Semi-Quantitative analysis (300ng/mL cutoff)

Concentration	% of cutoff	Result
0	-100%	80 negative
75	-75%	80 negative
150	-50%	80 negative
225	-25%	80 negative
300	Cutoff	38 negative 42 positive
375	+25%	80 positive
450	+50%	80 positive
525	+75%	80 positive
600	+100%	80 positive

b. *Linearity/assay reportable range:*
A drug free urine pool was spiked with a high concentration of Oxycodone and was used as the high value specimen. Additional pools were made by serially diluting the high value specimen with drug free urine in increments of 10%. Aliquots from each pool were analyzed in duplicate in the semi-quantitative mode using all 6 calibrators for the 100 ng/mL and 300 ng/mL cutoffs (0, 100, 300, 500, 1000 ng/mL) using 2 Beckman Coulter AU400e Chemistry Analyzers. For each known concentration, drug recovery was calculated using the mean concentration of the replicates. Summary results are listed below:

Expected Concentration (ng/mL)	Mean Measured Concentration (ng/mL)	Recovery (%)
25	27.4	110
50	50.2	100
100	100.4	100
200	228.1	114
300	307.3	102
400	443.8	111
500	514.1	103
600	606.6	101
700	732.9	105
800	787.1	98
900	870.9	104
1000	937	94
1100	969.8	88
1200	1001.4	83

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):* The controls and calibrators are prepared using a commercially available Oxycodone standard. The concentration of Oxycodone in these calibrators and controls is determined by LC/MS analysis.

Stability: Protocols and acceptance criteria were reviewed and found to be acceptable. The sponsor claims that when stored at 2-8 ºC calibrators and controls are stable for 10 months.

The sponsor claims that once opened, the calibrators and controls are stable for 14 days if stored on-board the instrument at ambient temperature (22 to 28ºC).

d. *Detection limit:*
Not applicable

e. *Analytical specificity:*

Interference studies:
Structurally non-similar compounds and endogenous compound were evaluated to ensure that there was no interference that caused a false response relative to the cutoff in the qualitative mode and semi-quantitative mode. Results were analyzed following the recommendations in CLSI EP 7-A2. Potential interfering and endogenous substances were spiked into drug free urine containing Oxycodone at ± 25% of the cutoff (75ng/mL and 125ng/mL for the 100ng/mL cutoff and 225ng/mL and 375ng/mL for the 300ng/mL cutoff). Each sample was tested in replicates of 4 for the qualitative testing and in duplicate for the semi-quantitative testing using 2 Beckman Coulter AU400e Chemistry Analyzers and compared to the corresponding Oxycodone control.

The following structurally non-similar compounds (each tested at 100,000 ng/mL) were found not to interfere with the test result at either cutoff in both qualitative and semi-quantitative modes:

Acetaminophen, Alprazolam, d-Amphetamine, Amitryptyline, Amobarbital, Benzoylecgonine, Bromazepam, Caffeine, Clonazepam, Carbamazine, Carisoprodol, Chlorpromazine, Desipramine, Dextromethorphan, Diazepam, Diphenhydramine, Doxepine, Doxylamine, Flunitrazepam, Flurazepam, Fluoxetine, Ibuprofen, Imipramine, Ketamine, Lidocaine, LSD, Lorazepam, 3,4-MDA, 3,4-MDEA, 3,4-MDMA, PMA, Medazepam, Methadone Metabolite (EDDP), Methaqualone, d-Methamphetamine, Meprobamate, Mephenytoin, Methylphenidate, Methadone, Naproxen, Nordiazepine, Nortriptyline, Oxazepam, Phenobarbital, Phencyclidine (PCP), Pentobarbital, Phenothiazine, Propoxyphene, Pentazocine, Protriptyline, Salicylic acid,

Secobarbital, Sertraline, Pseudo-ephedrine, Ranitidine, Temazepam, 11-nor-carboxy-Δ9-THC, Tramadol, Triazolam, Zolpidem

The following endogenous substances at the tested concentrations did not interfere with the results of the assay at either cutoff:

Compound	Concentration Tested
Acetone	1.0 g/dL
Ascorbic Acid	1.5 g/dL
Bilirubin	0.002 g/dL
Creatinine	0.5 g/dL
Ethanol	1.0 g/dL
γ-Globulin	0.5 g/dL
Glucose	2.0 g/dL
Hemoglobin	0.300 g/dL
Human Serum Albumin	0.5 g/dL
Oxalic Acid	0.1 g/dL
Riboflavin	0.0075 g/dL
Sodium Azide	1% w/v
Sodium Chloride	6.0 g/dL
Sodium Fluoride	1% w/v
Urea	6.0 g/dL

Boric acid at 1% w/v resulted in a false negative results at both cutoffs. The sponsor included the following limitation in the labeling:
"Boric acid at 1% w/v resulted in false negative results at both cutoffs. Boric Acid is not recommended as a preservative for urine."

Effect of pH: The sponsor evaluated the effect of pH on the test results using both qualitative and semi-quantitative modes. Drug free urine containing Oxycodone at ± 25% of the cutoff (75ng/mL and 125ng/mL for the 100ng/mL cutoff and 225ng/mL and 375ng/mL for the 300ng/mL cutoff) were pH adjusted using hydrochloric acid or sodium hydroxide. pH values of 3.0, 4.0, 5.0, 6.0, 7.0, 8.0, 9.0, 10.0 and 11.0 did not interfere with the test result at either cutoff.

Effect of specific gravity: The sponsor evaluated the effect of specific gravity on the test results using both qualitative and semi-quantitative modes. Drug free urine containing Oxycodone at ± 25% of the cutoff (75ng/mL and 125ng/mL for the 100ng/mL cutoff and 225ng/mL and 375ng/mL for the 300ng/mL cutoff) were adjusted using salt/albumin. Specific Gravity values of 1.000, 1.002, 1.005, 1.010, 1,015, 1.020, 1.025 and 1.030 did not interfere with the test result at either cutoff.

Cross reactivity from structurally related compounds was evaluated in the qualitative and semi-quantitative modes. Oxycodone and the structurally

similar compound (listed below) were spiked into drug free urine at levels that will trigger the 100ng and 300ng/mL Oxycodone cutoff. Each sample was tested in singlicate for the semi-quantitative mode and in replicates of 4 for the qualitative mode using 2 Beckman Coulter AU400e Chemistry Analyzers. The results are summarized below and are expressed as the minimum concentration of metabolite or compound required to produce a response approximately equivalent to each cutoff concentration of the assay. If no cross-reactivity was observed, then the concentration in the table is the concentration tested and "N.D." or none detected is reported.

100 ng/mL cutoff

Compound	Concentration Tested (ng/mL)	Cross-Reactivity (%)
Oxymorphone	100	100
Noroxymorphone	5,000	2.00
Oxymorphone-3β-Glucuronide	500	20.00
Noroxycodone	7,500	1.33
Naloxone	3,750	2.67
Naloxone, 3-Glucuronide	50,000	0.20
Naltrexone	30,000	0.33
Morphine	350,000	<0.10
Normorphine	1,000,000	N.D.
Codeine	500,000	N.D.
Dihydrocodeine	100,000	N.D.
Norcodeine	1,000,000	N.D.
Heroin	300,000	N.D.
Hydromorphone	50,000	N.D.
Hydrocodone	100,000	N.D.
Meperidine	50,000	N.D.
Levorphanol	200,000	N.D.
Buprenorphine	10,000	N.D.
Morphine-3β-Glucuronide	500,000	N.D.
6-Acetyl Morphine	100,000	N.D.

300 ng/mL cutoff

Compound	Concentration Tested (ng/mL)	Cross-Reactivity (%)
Oxymorphone	300	100
Noroxymorphone	50,000	0.60
Oxymorphone-3β-Glucuronide	4,000	7.50
Noroxycodone	75,000	0.40
Naloxone	37,500	0.80

Naloxone, 3-Glucuronide	500,000	<0.10
Naltrexone	300,000	0.10
Morphine	350,000	N.D.
Normorphine	1,000,000	N.D.
Codeine	500,000	N.D.
Dihydrocodeine	100,000	N.D.
Norcodeine	1,000,000	N.D.
Heroin	300,000	N.D.
Hydromorphone	50,000	N.D.
Hydrocodone	100,000	N.D.
Meperidine	50,000	N.D.
Levorphanol	200,000	N.D.
Buprenorphine	10,000	N.D.
Morphine-3β-Glucuronide	500,000	N.D.
6-Acetyl Morphine	100,000	N.D.

f. Assay cut-off:

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

2. Comparison studies:

a. Method comparison with predicate device:

The method comparison study was performed in-house using 169 unaltered, leftover clinical urine samples obtained from clinical testing laboratories and were analyzed using one lot of the proposed device using 2 Beckman Coulter AU400e Chemistry Analyzers. The sponsor compared the results of their device to results obtained using liquid chromatography/mass spectroscopy (LC/MS). The results of the assay performance compared to LC/MS are summarized below:

The following table summarizes the performance of the assay for the 100ng/mL cutoff:

100 ng/mL cutoff

Candidate Device Result	Oxycodone Concentration (ng/mL)					Agreement (%)
	NEG	<50	50-99	100-150	>150	
Qualitative/POS	0	0	0	11	78	100
Qualitative/NEG	63	8	9	0	0	100
Semi-quant/POS	0	0	0	11	78	100
Semi-quant/NEG	63	8	9	0	0	100

The following tables summarize the performance of the assay for the 300ng/mL cutoff:

300 ng/mL cutoff

Candidate Device Result	Oxycodone Concentration (ng/mL)					Agreement (%)
	NEG	<150	150-299	300-450	>450	
Qualitative/POS	0	2*	5**	15	67	100
Qualitative/NEG	43	27	10	0	0	92
Semi-quant/POS	0	2*	5**	15	67	100
Semi-quant/NEG	43	27	10	0	0	92

Discordant samples at the 300 ng/mL cutoff

Candidate Device Result				LC/MS Result (ng/mL)
Sample	Qualitative	Semi-Quantitative		
		Value	Result	
10478*	POS	479	POS	Oxycodone at 60 and Oxymorphone at 45
10203**	POS	407	POS	Oxycodone at 61 and Oxymorphone at 91
10466*	POS	431	POS	Oxycodone at 100 and Oxymorphone 38
10472**	POS	841	POS	Oxycodone at 157 and Oxymorphone at 11
10477**	POS	1409	POS	Oxycodone at 173 and Oxymorphone at 75
10192**	POS	603	POS	Oxycodone at 180 and Oxymorphone at 8
10471**	POS	553	POS	Oxycodone at 196 and Oxymorphone at 60

* The Sponsor investigated the cause of these false positive results and suspected sample integrity issues though this could not be verified. Therefore the Sponsor conducted testing of additional samples and the false results were not replicated in this study.

b. Matrix comparison:
This device is intended for use on 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.