

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

A. 510(k) Number:

k051577

B. Purpose for Submission:

New device

C. Measurand:

Methamphetamine

D. Type of Test:

Qualitative immunoassay

E. Applicant:

Immunalysis Corporation

F. Proprietary and Established Names:

Immunalysis Methamphetamine ELISA for Oral Fluids

G. Regulatory Information:

1. Regulation section:

21 CFR §862.3610, Methamphetamine Test System

2. Classification:

Class II

3. Product code:

LAF

4. Panel:

Toxicology (91)

H. Intended Use:

1. Intended use(s):

See indications for use below.

2. Indication(s) for use:

The Immunalysis Methamphetamine ELISA test system utilizes an Enzyme Linked Immunoassay (ELISA) for the qualitative detection of methamphetamine in ORAL FLUID SAMPLES COLLECTED WITH THE QUANTISAL™ ORAL FLUID COLLECTION DEVICE ONLY using a cutoff of 50 ng/mL of d-methamphetamine. This in vitro diagnostic device is intended for clinical laboratory use only.

The Immunalysis Methamphetamine ELISA Kit for Oral Fluids 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 (GS-MS) is the preferred confirmatory method (1). Clinical and Professional judgment should be applied to any drug of abuse test result, particularly when preliminary positive results are used.

3. Special conditions for use statement(s):

See above.

4. Special instrument requirements:

This device must be read on a spectrophotometer that reads wavelengths of 450 nm and 620 nm.

I. Device Description:

The device consists of a saliva collection device and a methamphetamine ELISA kit. An oral fluid specimen is collected by placing the collection device, a cellulose pad affixed to a propylene stem under the tongue, until approximately one milliliter saliva has saturated the pad. A blue indicator on the stem indicates when enough sample has been collected. The collector is transferred to a provided polypropylene tube containing preservative buffer (3 ml) and closed, ready for transport or storage. The ELISA assay consists of 8-well microstrips coated with high affinity purified rabbit polyclonal antibody, a plate frame, conjugated methamphetamine, negative and positive controls, a cut-off calibrator, TMB substrate, and stop reagent.

J. Substantial Equivalence Information:

1. Predicate device name(s):
DRI Amphetamines EIA Assay
2. Predicate 510(k) number(s):
k934891
3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Analyte	Methamphetamine	Methamphetamine and Amphetamine
Methodology	Immunoassay (EIA)	Immunoassay (ELISA)

Differences		
Item	Device	Predicate
Test Matrix	Oral Fluid	Urine
Cutoff	50 ng/mL	1000 ng/mL

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

None referenced.

L. Test Principle:

Enzyme-labeled drug and drug present in the sample compete for limited anti-methamphetamine antibody binding sites. Binding of the enzyme-labeled drug inhibits its reaction with the substrate, thereby influencing the rate of absorbance change measured by the instrument. The rate of absorbance change is proportional to the concentration of drug in the sample. Concentrations of controls and unknowns are calculated from the standard curve. Results are read at 450 and 620 nm.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:
 - a. *Precision/Reproducibility:*

Precision was tested by spiking negative oral fluid with 0, 25, 50, 75, and 100 ng/mL of d-methamphetamine; this corresponded to 0, 50%, 100%, 150%, and 200% of the cutoff. One milliliter of the spiked fluids were pipetted onto the collection pad of the oral fluid collector then processed as per instructions.

Intra-assay precision was assessed with sixteen replicates of each concentration analyzed in one run:

**Intra-assay Precision: Immunalysis Methamphetamine
for Oral Fluid Assay**

Methamphetamine (ng/mL)	Mean OD	Std Dev	CV
0	2.35	0.055	2.33 %
25 (50% c/o)	1.07	0.048	4.43 %
50 (100% c/o)	0.80	0.032	3.97 %
75 (150% c/o)	0.64	0.030	4.62 %
100 (200% c/o)	0.54	0.027	5.07 %

Inter-assay precision was assessed by eight replicates of each concentration run in 10 different assay runs (2 per day over 5 days). Results are expressed as B/B0% where B = absorbance of sample and B0 = absorbance of the zero calibrator.

**Inter-assay Precision: Immunalysis Methamphetamine
for Oral Fluid Assay**

	Methamphetamine Concentration (ng/mL)			
	25 (50% c/o)	50 (100% c/o)	75 (150% c/o)	100 (200% c/o)
Mean	44.2	31.7	26.5	23.1
Std.Dev	2.0	1.6	1.6	1.1
% CV	4.6	5.2	5.9	4.8

Reproducibility of the oral fluid collection device was assessed by collecting oral fluid from 50 subjects with a pre-weighed collector and tube as per the package instructions. After the volume indicator turned blue, the collector and tube were weighed and the net weight of the saliva was determined and converted to volume (mLs).

Quantisal Oral Fluid Collection Device: Volume Adequacy Study

Avg. Vol. (mL)	Std. Dev.	C.V.	Mean + 3 SD (mL)	Mean - 3 SD (mL)
0.993	0.029	2.88%	1.079	0.907

These results support the sponsor's claim that the device collects 1 mL \pm 10% saliva.

b. Linearity/assay reportable range:

Not applicable. This assay is intended for qualitative use.

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

All calibrator and control stock solutions are prepared from commercially available DEA-exempt solutions; d-methamphetamine concentration is confirmed by GC/MS analysis.

Claimed shelf-life of the oral fluid collection device and the ELISA kit and components is 24 months and 12 months respectively. Real-time studies are ongoing.

Stability of methamphetamine in the collection device was determined by spiking a pool of negative saliva with methamphetamine at a concentration around the cutoff and a pool at a concentration two-fold higher (2X). Samples were stored at 4°C or at room temperature. The specimens kept at room temperature were assayed in duplicate by GC-MS after 7 days, 14 days and 30 days; samples kept at 4°C were assayed were assayed in duplicate by GC-MS after 14 days and 30 days. The sponsor's acceptance criterion was recovery of \pm 20% of the initial value.

Stability of Methamphetamine in Quantisal Buffer

Stability at Room Temperature					
Day	Meth spike (ng/mL)	% initial value		2X Meth spike (ng/mL)	% initial value
0	40.03	100		81.90	100
7	41.30	103.2		80.34	98.1
14	41.57	103.8		72.70	88.77
21	39.04	97.53		76.12	92.94
30	38.40	95.93		69.24	84.54
Stability at 4°C					
0	40.03	100		81.90	100
14	40.00	99.9		69.40	84.7
30	36.71	91.7		74.08	90.5

A shipping study showed that methamphetamine spiked into saliva had acceptable recovery (\pm 15%) after transport.

d. Detection limit:

See the Precision/Reproducibility section above for performance around the stated cutoff concentration.

e. *Analytical specificity:*

Cross-reactivity of structurally similar compounds was determined by spiking concentrations of different drugs into synthetic oral fluid.

**Cross-reactivity with similar compounds:
Immunalysis Methamphetamine Oral Fluid Assay**

Compound	Conc. Tested (ng/mL)	Cross-Reactivity	Compound	Conc. Tested (ng/mL)	Cross-Reactivity
d-methamphetamine	50	100%	(+) pseudoephedrine	1000	3.7%
l-methamphetamine	1000	2%	(+) pseudoephedrine	5000	1.5%
d-amphetamine	1000	<1%	(-) pseudoephedrine	500	<1%
l-amphetamine	5000	<1%	phenolpropanolamide	10000	<1%
dl-amphetamine	1000	<1%	(+) ephedrine	5000	<1%
dl-MDMA	25	98%	(-) ephedrine	5000	2.3%
dl-MDMA	50	78%	fenfluramine	1000	1.8%
dl-MDA	1000	<1%	diphenhydramine	10000	<1%
dl-MDEA	250	6.4%			

Structurally unrelated compounds were spiked into synthetic oral fluid at a concentration of 10000 ng/mL; none of the compounds in the table below had an immunoassay response greater than an equivalent of 25 ng/mL d-methamphetamine.

**Compounds tested for Cross-reactivity:
Immunalysis Methamphetamine Oral Fluid Assay**

Acetaminophen	Ethylmorphine	Mereridine
Amitriptyline	Flurazepam	Nalorphine
Amobarbital	Glutethimide	Nicotine
Barbital	Hexobarbital	Nordoxepin
Benzoylecgonine	Hydromorphone	n-Normethsuximide
Butabarbital	Imipramine	Nortriptyline
Bromazepam	Lidocaine	Oxazepam
Caffeine	Lorazepam	Oxycodone
Carbamazepine	Medazepam	Phenobarbital
Cocaine	Methadone	Phensuximide
Codeine	EDDP	Phenytoin
Chlorpromazine	Methaqualone	Primidone
Desipramine	Metharbital	Protriptyline
Diacetylmorphine	Mephenytoin	Quinine
Dihydrocarbamazepine	Methyl-propylsuccinimide	Secobarbital
Diazepam	Mephobarbital	Temazepam
Doxepin	Methyl PEMA	Theophylline
Dyphylline	Methsuximide	Trimipramine
Ethosuximide	4-Methylprimidone	
Ethotoin	Morphine	

Commonly ingested substances were tested for interference. Sugar 145 mg/mL, toothpaste 25 mg/mL, cranberry juice 25% v/v, baking soda 25

mg/mL, orange juice 25% v/v, carbonated cola 25% v/v, cough syrup 10% v/v, mouthwash 25% v/v, distilled water. Substances were diluted or dissolved in distilled water then spiked with 25 ng/mL methamphetamine (50% cutoff) or 75 ng/mL methamphetamine (150% cutoff) and tested. Results were compared to the cutoff calibrator to determine if the sample was positive or negative; specific effects were compared to the same methamphetamine concentration in synthetic oral fluid:

**Effect of Common Compounds on
Immunalysis Methamphetamine Oral Fluid Assay**

Compound	Mean Abs	B/B0%	POS/NEG	Mean Abs	B/B0%	POS/NEG
Synthetic Oral Fluid	2.834	100				
25 ng/mL methamphetamine	1.347	47.5	NEG			
50 ng/mL methamphetamine	0.952	33.6	Cutoff			
100 ng/mL methamphetamine	0.627	22.1	POS			
	25 ng/mL Methamp spike			75 ng/mL Methamp spike		
Distilled water	1.612	56.9	NEG	0.861	30.4	POS
Sugar water sol'n	1.454	51.3	NEG	0.808	28.5	POS
Toothpaste slurry	1.334	47.1	NEG	0.784	27.6	POS
Cranberry juice	1.448	51.1	NEG	0.834	29.4	POS
Baking Soda sol'n	1.501	53.0	NEG	0.919	32.4	POS
Orange juice	0.944	33.3	NEG	0.689	24.3	POS
Cola	1.347	47.5	NEG	0.844	29.8	POS
Cough syrup*	0.193	6.81	POS	0.165	5.81	POS
Mouthwash	1.528	53.9	NEG	0.836	29.5	POS

* Cough syrup contained 2 mg/mL (+) pseudoephedrine, sufficient to cause cross-reactivity in the assay. See cross-reactivity with related compounds table above.

f. Assay cut-off:

Performance around the assay cut-off of 50 ng/mL is demonstrated in the intra-assay precision section above.

The Substance Abuse and Mental Health Services Administration (SAMHSA) has recommended 50 ng/mL as a cutoff level for methamphetamine oral fluid tests.

2. Comparison studies:

a. Method comparison with predicate device:

Oral fluid and urine samples were collected in the same visit from 185 admitted methamphetamine users in a clinical setting. Urine samples were tested by the predicate assay using a cutoff of 1000 ng/mL. Oral fluid samples

were tested in duplicate using a screening cutoff of 50 ng/mL; all samples were tested by GC/MS at an independent facility. Thirteen samples were $\pm 50\%$ of the cutoff (by GC/MS).

Comparison of Immunalysis Methamphetamine Oral Fluid Assay and the Predicate Urine Assay

		Predicate Urine Assay	
		Pos	Neg
Methamphetamine Oral Fluid Assay	Pos	62	6
	Neg	10	107

Positive agreement: 91 %
 Negative agreement: 92 %
 Overall agreement: 91 %

Comparison of Immunalysis Methamphetamine Oral Fluid Assay and GC/MS

		GC/MS	
		Pos	Neg
Methamphetamine Oral Fluid Assay	Pos	66	2
	Neg	0	117

Positive agreement: 97 %
 Negative agreement: 100 %
 Overall agreement: 99 %

b. Matrix comparison:

Not applicable; this device is intended for use with oral fluid 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):

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.