

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

A. 510(k) Number:

k062637

B. Purpose for Submission:

New product

C. Measurand:

Methadone

D. Type of Test:

Semi-quantitative and qualitative homogeneous enzyme immunoassay

E. Applicant:

Ortho-Clinical Diagnostic

F. Proprietary and Established Names:

VITROS Chemistry Products METD Reagent

VITROS Chemistry Products Calibrator Kit 26

VITROS Chemistry Products DAT Performance Verifiers I, II, III, IV, and V

G. Regulatory Information:

1. Regulation section:

21 CFR §862.3620, Methadone Test System

21 CFR §862.3200, Clinical Toxicology Calibrator

21 CFR §862.3280, Clinical Toxicology Control Material

2. Classification:

Class II (Reagent, Calibrator)

Class I, Reserved (Control)

3. Product code:

DJR; DKB; DIF

4. Panel:

Toxicology (91)

H. Intended Use:

1. Intended use(s):

See Indications for Use below.

2. Indication(s) for use:

VITROS Chemistry Products METD Reagent is used on VITROS 5,1 FS Chemistry Systems for the semi-quantitative or qualitative determination of methadone (METD) in human urine using a cutoff of either 150 ng/mL or 300 ng/mL. Measurements obtained with the VITROS METD method are used in the diagnosis and treatment of methadone use or overdose.

The VITROS Chemistry Products METD assay is intended for use by professional laboratory personnel. It provides only a preliminary test result. A more specific alternative chemical method must be used to confirm a result obtained with this assay. Gas chromatography/mass spectrometry (GC/MS) is the preferred confirmatory method. Clinical consideration and professional judgment should be applied to any drug-of-abuse test result, particularly when evaluating a preliminary positive result.

VITROS Chemistry Products Calibrator Kit 26 is used to calibrate VITROS 5.1 FS Chemistry Systems for the qualitative or semi-quantitative measurement of drugs of abuse.

VITROS Chemistry Products DAT Performance Verifiers are assayed controls used to monitor performance of urine drugs of abuse screening assays on VITROS 5,1 FS Chemistry Systems.

3. Special conditions for use statement(s):

This device is for use by professional laboratory personnel. For *in vitro* diagnostic use only.

The VITROS Chemistry Products METD assay is intended for use by professional laboratory personnel. It provides only a preliminary test result. A more specific alternative chemical method must be used to confirm a result obtained with this assay. Gas chromatography/mass spectrometry (GC/MS) is the preferred confirmatory method. Clinical consideration and professional judgment should be applied to any drug-of-abuse test result, particularly when evaluating a preliminary positive result.

4. Special instrument requirements:

Ortho-Clinical Diagnostics VITROS 5,1 FS Chemistry System

I. Device Description:

The VITROS METD Reagent is a dual chambered package containing ready-to-use liquid reagents that are used to detect methadone in urine. Sample, calibrators, and controls are automatically treated with surfactant (DAT Diluent 2) prior to addition of reagents. Treated sample is added to Reagent 1 containing antibodies reactive to methadone, glucose-6-phosphate and nicotinamide adenine dinucleotide (NAD⁺), followed by Reagent 2 containing methadone labeled with the enzyme glucose-6-phosphate dehydrogenase (G6P-DH).

VITROS Chemistry Products Calibrator Kit 26 is prepared from human urine to which drugs of abuse, metabolites of drugs of abuse, organic salts, surfactants and preservative have been added.

VITROS DAT Performance Verifiers I, II, III, IV & V are prepared from a human urine pool to which analytes, surfactant and preservative have been added. These are assayed controls used to monitor performance of the VITROS METD Reagent on VITROS 5,1 FS Chemistry Systems.

The product labeling for the Calibrator Kit 26 and Performance Verifiers contain warnings regarding the presence of human sourced materials and recommend the use

of Universal Precautions when handling these products.

J. Substantial Equivalence Information:

1. Predicate device name(s):
EMIT II Plus Methadone Assay
Liquicheck Urine Toxicology Controls
2. Predicate 510(k) number(s):
k010962; k022707
3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	For use in the qualitative and semi-quantitative analysis of methadone in human urine.	Same
Reagent	Liquid, ready to use	Same
Principle	Homogeneous enzyme immunoassay	Same
Matrix	Urine	Same
Antibody	Sheep polyclonal	Same

Differences		
Item	Device	Predicate
Instrumentation	VITROS 5,1 FS Chemistry Systems	Multiple automated clinical chemistry analyzers
Calibrators	Six levels	Four levels
Controls	Five levels	Two levels

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

CLSI EP9-A2: Method Comparison and Bias Estimation Using Patient Samples
CLSI EP5-A: Evaluation of Precision Performance of Clinical Chemistry Devices
CLSI EP6-A: Evaluation of the Linearity of Quantitative Measurement Procedures, A Statistical Approach
CLSI EP7-P: Interference Testing in Clinical Chemistry
CLSI EP17-A: Protocols for Demonstration, Verification and Evaluation of Limits of Detection and Quantitation
CLSI EP12-A: User Protocols for Evaluation of Qualitative Test Performance

L. Test Principle:

The VITROS METD Reagent is a dual chambered package containing ready-to-use liquid reagents that are used to detect methadone in urine. Sample, calibrators, and controls are automatically treated with surfactant (DAT Diluent 2) prior to addition of reagents. Treated sample is added to Reagent 1 containing antibodies reactive to methadone, glucose-6-phosphate and nicotinamide adenine dinucleotide (NAD⁺),

followed by Reagent 2 containing methadone labeled with the enzyme glucose-6-phosphate dehydrogenase (G6P-DH). The assay is based on competition between methadone in the treated urine sample and the methadone labeled with the enzyme glucose-6-phosphate dehydrogenase (G6P-DH) for antibody binding sites. Enzyme activity decreases upon binding to the antibody, therefore the concentration of methadone in the urine sample is directly proportional to measured enzyme activity. Active enzyme converts oxidized nicotinamide adenine dinucleotide (NAD⁺) to NADH, resulting in an absorbance change that is measured spectrophotometrically at 340 nm.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

Precision was evaluated with human urine-based quality control materials on the VITROS 5,1 FS Chemistry System following CLSI Protocol EP5 and CLSI protocol EP12.

Semi-Quantitative

System	Conventional Units (ng/mL) and SI Units (µg/L)			Within Lab CV%**	No. Observ.	No. Days
	Mean Conc.	Within Day SD*	Within Lab SD**			
VITROS 5,1 FS	108	5.1	6.2	5.7%	88	22
	149	5.7	6.8	4.6%	87	22
	190	5.2	6.5	3.4%	88	22
	233	4.8	6.6	2.8%	88	22
	312	4.0	4.5	1.4%	88	22
	377	4.0	5.6	1.5%	88	22
	624	17.3	22.8	3.7%	88	22

* Within Day imprecision was determined using two runs per day with two replications per run.

** Within Lab imprecision was determined using a single lot of reagents with one analyzer and five calibrations.

Qualitative imprecision was assessed using test fluids targeted at $\pm 25\%$ of each cutoff. The imprecision was determined as the confidence level of obtaining a correct result with known positive or negative fluids.

Qualitative*

System	Cutoff Level (ng/mL & µg/L)	Test Fluid at $\pm 25\%$ Cutoff	Number of Observation s	Number of Correct Interpretations	Confidence Level
VITROS 5,1 FS	150	-25%	88	88	>95% negative reading
	150	+25%	88	88	>95% positive reading
	300	-25%	88	88	>95% negative reading

Qualitative*

System	Cutoff Level (ng/mL & □g/L)	Test Fluid at ± 25% Cutoff	Number of Observation s	Number of Correct Interpretations	Confidence Level
	300	+25%	88	88	>95% positive reading

* Determined using two runs per day with two replicates per run for 22 days, using a single lot of reagents with one analyzer and five calibrations

b. Linearity/assay reportable range:

The sponsor followed CLSI EP6-A in determining the linear range of their device. Two urine pools were prepared with methadone concentrations at the low (0 ng/mL) and high (1000 ng/mL) end of the calibration range. The two pools were mixed to give 14 admixtures of intermediate concentrations. Linearity was evaluated using two assay reagent lots and comparing the measured results against the expected results from 16 pooled samples. A linear regression was performed and the results indicated acceptable linearity across the methadone concentration range tested (12 to 736 ng/mL). The reportable range of the VITROS METD assay is 40-735 ng/mL.

Recovery of Semi-Quantitative Results

Eleven admixtures spanning the reportable range (40 to 735 ng/mL) were prepared from two human urine-based pools. Methadone values for the admixtures were calculated based on gravimetric addition of methadone with GC/MS verification of the high and low pools. Percent recovery was calculated using the concentration obtained by the VITROS Chemistry Products METD Assay versus the nominal methadone value.

Recovery of Methadone

Nominal Methadone Concentration (ng/mL)	VITROS METD Assay (ng/mL)	% Recovery
56	53	94.8
75	74	99.3
100	98	98.4
150	150	100.0
200	204	102.2
250	255	101.9
300	297	99.0
375	370	98.8
500	489	97.8
625	632	101.1
700	693	99.1

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

The assigned values for the calibrators and controls are traceable to the Cerilliant methadone standard catalogue M-110 and are verified by GC/MS. Real time and accelerated stability studies were conducted; protocols and acceptance criteria were described and found to be acceptable. These studies support the manufacturer's stability claims. Real time studies are ongoing

d. *Detection limit:*

The detection limit was determined according to protocol recommendations in CLSI EP-17. The claimed lower limit for VITROS METD assay is 40 ng/mL.

e. *Analytical specificity:*

The specificity of the VITROS Chemistry Products METD assay for various methadone metabolites and structurally similar compounds was estimated by generating a dose-response curve for each of the compounds listed below. The quantity (ng/mL) of a compound necessary to produce a value equivalent to the methadone quantity (ng/mL) at each cutoff value is listed below. If a sample contains more than one compound detected by the assay, lower quantities of cross-reactant than those listed below may produce a value approximately equivalent to or greater than that of the cutoff value due to their combined effects.

Substances that Cross-react with METD

Compound	Quantity (ng/mL) equivalent to 150 ng/mL of methadone	% cross-reactivity *	Quantity (ng/mL) equivalent to 300 ng/mL of methadone	% cross-reactivity *
L-A-methadol	302	49.67%	2625	11.43%
LAMM	385	38.96%	5400	5.56%
thioridazine	54,000	0.28%	>300,000	<0.1%
EDDP	171,000	0.09%	>300,000	<0.1%
sertraline	300,000	0.05%	>1,000,000	<0.03%
EMDP	>300,000	<0.05%	>300,000	<0.1%

* The VITROS METD Assay cutoff value (ng/mL) divided by the amount of cross-reactant (ng/mL) that produces a value equivalent to the cutoff value, multiplied by 100.

The substances listed in the table, at the concentrations shown, were tested according to CLSI Protocol EP7 and found not to interfere, bias <28 ng/mL at 150 ng/mL methadone and bias <57 ng/mL at 300 ng/mL methadone.

Substances that Do Not Interfere with METD

Compound	Concentration	
ammonia	570 mg/dL	316 mmol/L
ascorbic acid	500 mg/dL	28.4 mmol/L
bilirubin	26 mg/dL	444 µmol/L
calcium	30 mg/dL	7.5 mmol/L

Substances that Do Not Interfere with METD

Compound	Concentration	
ciprofloxacin	10 mg/dL	302 µmol/L
citric acid	100 mg/dL	5.2 mmol/L
cloxacillin	10 mg/dL	230 µmol/L
creatinine	300 mg/dL	26.5 mmol/L
diethylpropione	10 mg/dL	487 µmol/L
ethacrynic acid	10 mg/dL	330 µmol/L
ethanol	780 mg/dL	169 mmol/L
glucose	4000 mg/dL	222 mmol/L
hemoglobin	500 mg/dL	5.00 g/L
human igg	200 mg/dL	2.00 g/L
human serum albumin	200 mg/dL	2.00 g/L
indomethacin	10 mg/dL	279 µmol/L
iron	0.1 mg/dL	17.9 µmol/L
potassium	587 mg/dL	150 mmol/L
magnesium	60 mg/dL	24.7 mmol/L
metronidazole	10 mg/dL	584 µmol/L
nylidrine	10 mg/dL	334 µmol/L
oxalic acid	300 mg/dL	23.8 mmol/L
ph = 4		
ph= 9		
phenylbutazone	10 mg/dL	324 µmol/L
phosphate	950 mg/dL	100 mmol/L
pyruvate	200 mg/dL	22.8 mmol/L
ranitidine hcl	10 mg/dL	318 µmol/L
riboflavin	2 mg/dL	53 µmol/L
theophylline	10 mg/dL	555 µmol/L
tolmetin/tolectin	10 mg/dL	390 µmol/L
trimethobenzamide hcl	10 mg/dL	257 µmol/L
tyramine	10 mg/dL	576 µmol/L
urea	3000 mg/dL	499.5 mmol/L
uric acid	120 mg/dL	7.14 mmol/L

f. Assay cut-off:

The stated cutoff of this assay is either 150ng/mL or 300 ng/mL.

2. Comparison studies:

a. Method comparison with predicate device:

A total of 140 human urine samples were assayed using the VITROS Chemistry Products METD Reagent and a commercially available immunoassay method. Percent agreement was evaluated at assay cutoffs of 150 and 300 ng/mL.

To challenge performance at the 150 ng/mL cutoff value, 43 of the 140 samples tested had concentrations within +/- 50% of the cutoff value.

To challenge performance at the 300 ng/mL cutoff value, 56 of the 140 samples tested had concentrations within +/- 50% of the cutoff value.

Predicate Device Comparison - METD

Cutoff Value (ng/mL)		Commercial Negative	Commercial Positive	% Agreement		
				% Agreement Negative	% Agreement Positive	% Agreement Overall
150	VITROS Positive	1*	91	98.0	100	99.3
	VITROS Negative	48	0			
300	VITROS Positive	1*	48	98.9	100	99.3
	VITROS Negative	91	0			

*See Commercial Method Summary of Discordant Results below.

Summary of Discordant Results: Commercial Method

Cutoff Value (ng/mL)	VITROS METD Assay (ng/mL)	Commercial Method (ng/mL)
150	199	149
300	310	291

A total of 139 human urine samples were assayed using the VITROS Chemistry Products METD Reagent and a GC/MS reference method for methadone. Percent agreement was evaluated at assay cutoff values of 150 and 300 ng/mL.

To challenge performance at the 150 ng/mL cutoff value, 59 of the 139 samples tested had concentrations within +/- 50% of the cutoff value.

To challenge performance at the 300 ng/mL cutoff value, 42 of the 139 samples tested had concentrations within +/- 50% of the cutoff value.

GC/MS Reference Method Comparison for METD

Cutoff Value (ng/mL)		GC/MS Reference Method				% Agreement		
		Low Negative	Near Cutoff Negative	Near Cutoff Positive	High Positive	% Agreement Negative	% Agreement Positive	% Agreement Overall
150		(<-50%) <75 ng/mL	(-50% to cutoff) 75-150 ng/mL	(cutoff to +50%) 150-225 ng/mL	(>+50%) >225 ng/mL	64.0	100	80.6
	VITROS Positive	1*	26*	18	46			
	VITROS Negative	33	15	0	0			

GC/MS Reference Method Comparison for METD

		GC/MS Reference Method				% Agreement		
Cutoff Value (ng/mL)		Low Negative	Near Cutoff Negative	Near Cutoff Positive	High Positive	% Agreement Negative	% Agreement Positive	% Agreement Overall
300		(<-50%) <150 ng/mL	(-50% to cutoff) 150-300 ng/mL	(cutoff to +50%) 300-450 ng/mL	(>+50%) >450 ng/mL	87.5	100	90.6
	VITROS Positive	1*	12*	13	22			
	VITROS Negative	74	17	0	0			

*See GC/MS Summary of Discordant Results below

Summary of Discordant Results: GC/MS			
Cutoff Value (ng/mL)	VITROS METD (ng/mL)	GC/MS (ng/mL)	Major Drug Present by GC/MS
150	151	136	Methadone
	156	128	
	160	141	
	162	125	
	166	83	
	170	115	
	171	132	
	178	115	
	181	146	
	181	149	
	182	108	
	199	124	
	199	<RR	
	211	125	
	228	130	
	231	137	
	231	143	
	238	119	
	246	82	
	248	127	
	249	88	
	261	139	
	265	102	
	271	119	
	271	122	
	277	136	
	310	112	
300	310	112	Methadone
	310	284	
	314	269	
	316	223	

Summary of Discordant Results: GC/MS			
Cutoff Value (ng/mL)	VITROS METD (ng/mL)	GC/MS (ng/mL)	Major Drug Present by GC/MS
	320	219	
	322	190	
	323	290	
	325	262	
	327	253	
	331	297	
	332	297	
	354	152	
	586	292	

- b. Matrix comparison:*
Not applicable; this device is for use with urine 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.