

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

A. 510(k) Number:

k041143

B. Purpose of the Submission:

New 510(k)

C. Analyte:

Barbiturate

D. Type of Test:

Qualitative immunoassay and associated calibrators

E. Applicant:

Randox Laboratories, Ltd.

F. Proprietary and Established Names:

Randox Barbiturate Assay

G. Regulatory Information:

1. Regulation section:

862.3150, Enzyme Immunoassay, Barbiturate

862.3200, Calibrator, Drug Mixture

2. Classification:

All class II

3. Product Code:

DIS and DKB, respectively

4. Panel:

Toxicology (91)

H. Intended Use:

1. Intended use(s):

Refer to Indications for use.

2. Indication(s) for use:

The evidence barbiturates test has been designed for use only on the evidence analyser for qualitative detection of barbiturates in urine using a cutoff concentration of 200 ng/mL Phenobarbital. Qualitative results obtained can be utilized in the diagnosis and treatment of Barbiturate use or overdose.

Evidence Drugs of Abuse Calibrators are liquid Calibrators containing phosphate buffer, preservatives and specified drug concentrations. There are nine levels of calibrator. They have been developed for the system.

3. Special condition for use statement(s):

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 is the preferred confirmatory method. Other chemical confirmation methods are available. Clinical consideration and professional judgment should be applied to any drug of abuse test result, particularly when preliminary positive results are used.

The assay is for Rx use.

The assay was not evaluated in point-of-care settings.

4. Special instrument Requirements:

The assay is for use only on the automated evidence Analyser, cleared under k030360. The originally cleared version of this calibrator was also included in k030360.

I. Device Description:

The evidence analyser is a fully automated Biochip Array System. It performs simultaneous detection of multiple analytes from a single patient sample. The core technology is the Randox Biochip, a solid-state device containing an array of discrete test regions containing immobilized antibodies specific to different drugs of abuse compound classes.

Calibrator EV3550 is a phosphate buffer based material with Phenobarbital added. It is a 9 level calibrator set which includes 9 different concentrations of Phenobarbital. Calibrations are run daily.

J. Substantial Equivalence Information:

1. Predicate device name(s):

CEDIA DAU Barbiturate Assay, Microgenics

2. Predicate K number(s):

k936030

3. Comparison with predicate:

Both devices are for the qualitative determination of the same analyte(s) in the same matrix, and utilize the same cutoff concentration. Both are analyzed on instruments. The candidate device utilized chemiluminescent technology utilizing biochip array technology whereas the predicate is analyzed on a spectrophotometric analyzer.

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

The sponsor referenced the NCCLS EP5-T2 Precision document and the NCCLS Interference document, EP7-A.

L. Test Principle:

A competitive chemiluminescent immunoassay is employed for the assay with the drug in the specimen and drug labelled with horseradish peroxidase (HRP) being in direct competition for the antibody binding sites. Increased levels of drug in a specimen will lead to reduced binding of drug labelled with HRP and thus a reduction in chemiluminescence being emitted. The light signal generated from each of the test regions on the biochip is detected using digital imaging technology and compared to that from a stored calibration curve. A normalized value is calculated as a percentage of the signal intensity emitted from the cut-off point on the calibration curve relative to the signal intensity emitted from the sample test region. Samples producing a response value greater than, or equal to, the response value of the calibrator cut-off are considered positive (normalized result ≥ 100). Samples producing a response value less than the response value of the calibrator cut-off are considered negative (normalized result < 100).

Description of the test antibody: polyclonal sheep antibody against barbiturates.

M. Performance Characteristics (if/when applicable):1. Analytical performance:a. *Precision/Reproducibility:*

Total imprecision data was determined at two different locations by assaying seven calibrators for 20 days, 2 runs per day in replicates of 2 (n=80) based on a cut-off of 300 ng/mL according to National Committee of Clinical Laboratory Standards (NCCLS) EP5-T2.

Specimen description: calibrator

Number of days: twenty

Replicates per day: Duplicates run twice a day

Lots of product used: one

Operator: manufacturer staff

Testing Facility: manufacturers facility

Results of the study are presented below:

Total imprecision							
Concentration (ng/mL Phenobarbital)	132.2	179.3	199.7	225.6	272	344.9	814.7
Site 1 *	81.7	95.0	95.8	98.1	111.9	119.0	189.2
Site 1 SD *	8.5	7.7	10.1	8.0	8.8	12.6	14.8
Site 1 % CV	10.4	8.1	10.5	8.1	7.9	10.6	7.8
Site 2 *	86.9	93.9	96.9	101.8	110.1	120.0	186.1
Site 2 SD *	7.3	7.6	6.3	8.8	10.8	16.0	11.6
Site 2 % CV	8.4	8.1	6.5	8.6	8.7	13.3	6.2

* Relative Light Units (rlu)

Results for Site 1 and Site 2 are expressed as normalized valued.

b. Linearity/assay reportable range:

Not applicable. The assay is for qualitative use. It does, however, include a series of 9 calibrators. A representative calibration curve appears in the Operator's Manual.

c. Traceability (controls, calibrators, or method):

Nine levels of Calibrators are provided separately. The sponsor recommends daily calibrations.

The sponsor indicates that a Master Lot of calibrators has been quantified for the component drugs of abuse in all 9 levels by assaying 4 replicates for each component on GC/MS. The values assigned to each lot are the mean of those measurements. The laboratory performing the analysis is certified to the College of American Pathologists. The Master Lot is stored at -80 °C and is used to assign concentrations to subsequent calibrator lots.

A minimum of 20 replicates from each subsequent lot are assayed and quantified by direct comparison to the mean values of a minimum of 20 replicate standard curves from the Master Lot of calibrators. Results are assigned by applying mean readings to these standard curves.

Table 13 of the original information received from the sponsor displays a calibration curve. It appears adequate, i.e., the curve is not flat.

Stability:

Stability studies are summarized for the calibrators. Aliquots of the calibrators were stored at -80 °C for reference purposes (the baseline) while the remainder were stored at 2-8 °C. At 26, 52, and 104 weeks

the calibrator values are compared to the values of calibrators stored at -80 °C.

Accelerated studies are conducted in the same manner, but involve comparing samples stored at 37 °C to those stored at 2-8 °C.

For both the real time and accelerated stability study, the following acceptance criteria for the studies are used:

The Relative Light Units (rlu), curve shape (B/B_0 , where B is the rlu for an individual calibrator level and B_0 is the rlu for the level 1 calibrator) and normalized values are examined. A stability of 1 year (at 2-8 °C) is assigned when the % difference in either % B/B_0 or normalized values between the -80 °C and the 2-8 °C is less than 10%

Open vial stability was also assessed for 14 days, using acceptance criteria of 10% when compared to the baseline.

d. Detection limit:

The sensitivity of the assay was established by analyzing 20 repeat determinations of a GC/MS verified negative urine sample. The mean normalized value was calculated and 2 standard deviations added. The resultant normalized value of 32 represents the lowest concentration of Phenobarbital which can be distinguished from the zero calibrator with a confidence level of 95%

e. Analytical specificity:

Specificity and cross-reactivity of the assay was assessed by comparing the standard curves from selected compounds to the standard curve of phenobarbital. Each compound was diluted in GC/MS verified negative urine to the concentrations specified. The concentration of the compound rendering a signal equivalent to phenobarbital at the cut-off concentration of the assay was determined. The percentage cross reactivities of those compounds are presented in Table 1.

Table 1. Cross reactivity of barbiturate compounds at the cut-off concentration.

Compound	% Cross Reactivity
Phenobarbital	100
Secobarbital	512
Pentobarbital	183
Amobarbital	84
Barbital	40
Butalbital	172
Alphenal	257
Butabarbital	429
Cyclopentobarbital	251
p-Hydroxyphenobarbital	69

Compounds failing to generate a positive response (normalized result <100) appear in Table 2. Compounds were evaluated up to the concentration listed.

Table 2. Concentrations of compounds failing to produce a positive result.

Compound	Concentration (ug/mL)
Oxazepam	500
Lorazepam	500
Temazepam	500
Alprazolam	500
A-OH-Alprazolam	500
Nordiazepam	500
Nitrazepam	500
Flunitrazepam	500
Clonazepam	500
11-nor-9-THC-COOH	10
6-Monoacetylmorphine	500
Benzoylcegonine	100
Codeine	500
d-Amphetamine	500
MDA	500
MDEA	500
MDMA	500
Methadone	500
Methamphetamine	500
Morphine	500
Morphine-3-glucuronide	500
Pentobarbital	500
Phencyclidine	500

No interference was observed for the assay from the compounds shown in Table 3 when added to urine. This study was run in accordance with methods outlined in the NCCLS interference document, EP7-A. Specific gravity and pH ranges were assessed using a dose-response series based on a 200 ng/mL phenobarbital cut-off in GC/MS verified negative urine. Sodium chloride and hydrochloric acid / sodium hydroxide were used to vary specific gravity and pH ranges respectively. Result differences of <10% between test and control were deemed acceptable.

Table 3. Interfering compounds eliciting a negative response using a 200 ng/mL cut-off.

Compound	Concentration tested (mg/dL)
Acetaminophen	1 mg/mL
Acetone	1000
Acetylsalicylic acid	1 mg/mL
Ascorbic acid	1500
Caffeine	1 mg/mL
Creatinine	500
Ethanol	1000
Galactose	10
globulin	500
Glucose	3000
Haemoglobin	300
Human serum albumin	500
Ibuprofen	40
Oxalic acid	100
Ranitidine	90
Riboflavin	7.5
Sodium chloride	6000
Urea	3500
pH	Acceptable range 3.0 – 11.0
Specific gravity	Acceptable range 1.002 – 1.04 g/mL

f. Assay cut-off:

The identified cutoff concentration of the assay is standard for the industry. Characterization of how the device performs analytically around the claimed cutoff concentration was performed.

2. Comparison studies:

a. Method comparison with predicate device:

1334 urine samples were randomly collected and assayed with the **evidence** Barbiturate assay on the **Randox evidence** analyser by an independent laboratory using a comparative enzyme

immunoassay method followed by GC/MS confirmation of barbiturates for borderline samples, positive samples or where discrepancies between methods occurred. Total GC/MS quantities were calculated as the combined concentrations (added equally and unweighted) of phenobarbital, secobarbital, pentobarbital and butalbital.

Comparison of evidence[®] with competitor EIA

		Comparative EIA 200 ng/mL cut-off	
		+	-
Candidate Device	+	195	6 ^a
	-	1 ^b	1132

^aAll samples tested by GC/MS and 2 found to contain barbiturates below 200 mg/mL

^bSample tested by GC/MS and found to contain barbiturates below 200 ng/mL

Comparison of evidence[®] to GC/MS

		GC/MS 200 ng/mL cut-off	
		+	-
Candidate Device	+	164	30 ^c
	-	0	13

^cAll samples tested by GC/MS and fifteen found to contain barbiturate below 200 ng/mL

Comparison of evidence[®] to GC/MS

GC/MS quantified	evidence [®]	
	Positive	Negative
Negative (<75% cut-off)	20	12
Near cut-off (75-100% cut-off) ^a	10	1
Near cut-off positive (100-125% cut-off) ^b	16	0
Positive (>125% cut-off)	148	0
% agreement with GC/MS	85.5%	

^a All 11 sample GC/MS confirmed to contain barbiturates between 150-200 ng/mL

^b All 16 sample GC/MS confirmed to contain barbiturates between 200-250 ng/mL

b. Matrix comparison:

Not applicable. The assay is intended for only one sample matrix.

3. Clinical studies:

a. Clinical sensitivity:

Not applicable. Clinical studies are not typically submitted for this device type.

b. Clinical specificity:

Not applicable. Clinical studies are not typically submitted for this device type.

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. Conclusion:

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