

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

A. 510(k) Number:

k063131

B. Purpose for Submission:

New device

C. Measurand:

Carbamazepine and Valproic Acid

D. Type of Test:

Quantitative, homogenous enzyme immunoassay

E. Applicant:

Thermo Fisher Scientific Oy

F. Proprietary and Established Names:

Carbamazepine

Valproic Acid

TDM Calibration Set B

G. Regulatory Information:

1. Regulation section:

21 CFR 862.3645, Neuroleptic drugs radioreceptor assay test system.

(Carbamazepine and Valproic Acid)

21 CFR 862.3200, Calibrator Drug Mixture

2. Classification:

II

3. Product code:

KLT, Enzyme Immunoassay, Carbamazepine

LEG, Enzyme Immunoassay, Valproic Acid

DKB, Clinical Toxicology Calibrator

4. Panel:

Toxicology (91)

H. Intended Use:

1. Intended use(s):

See Indications for use.

2. Indication(s) for use:

Carbamazepine

The Carbamazepine is intended for the quantitative *in-vitro* diagnostic determination of the carbamazepine concentration in human serum using T60 Clinical Chemistry Analyzers. Measurements are used in the diagnosis and treatment of carbamazepine overdose and in monitoring levels of carbamazepine to help ensure proper therapy.

Valproic Acid

The Valproic Acid is intended for quantitative *in-vitro* diagnostic determination of the valproic acid concentration in human serum using T60 Clinical Chemistry Analyzers. Measurements are used in the diagnosis and treatment of valproic acid

overdose and in monitoring levels of valproic acid to help ensure proper therapy.

Calibrator

TDM Calibration set B is intended for in vitro diagnostic use as a calibrator in the quantitative measurement of the kit code 981645 Carbamazepine and kit code 981650 Valproic acid assays on T60 Analyzer.

3. Special conditions for use statement(s):

Prescription use

4. Special instrument requirements:

T60 Clinical Chemistry Analyzers

I. Device Description:

Carbamazepine

The device consists of 4 reagents (Reagents A and B: buffer and lyophilizate).

Reagent A:

- buffer contains enzyme acceptor MOPS (3-(N-morpholino) propane sulfonic acid buffer, mouse monoclonal anti-carbamazepine antibodies, and NaN_3 .
- lyophilizate contains enzyme acceptor (microbial), buffer salts and NaN_3 .

Reagent B:

- buffer contains enzyme donor reconstitution MES (2-(N-morpholino) ethane sulfonic acid buffer and NaN_3 .
- lyophilizate contains enzyme donor (microbial) conjugated to carbamazepine, Chlorophenol red- β -D-galactopyranoside, buffer salts, and NaN_3 .

Valproic Acid

The device consists of 4 reagents (Reagents A and B: buffer and lyophilizate)

Reagent A:

- buffer contains enzyme acceptor HEPES - (N-[2-Hydroxyethyl] piperazine-N-[2-ethanesulfonic acid]) buffer) and NaN_3 .
- lyophilizate contains enzyme acceptor (microbial), mouse monoclonal anti- valproic antibody, BSA, sodium salicylate, buffer salts, and NaN_3 .

Reagent B:

- buffer contains enzyme donor reconstitution HEPES - (N-[2-Hydroxyethyl]piperazine-N-[2-ethanesulfonic acid]) buffer) and NaN_3 .
- lyophilizate contains enzyme donor (microbial), conjugated to valproic acid, Chlorophenol red- β -D-galactopyranoside, goat anti-mouse antibodies, buffer salts, and NaN_3 .

Calibrator

TDM Calibration set B consists of two levels of ready to use calibrators. The kit contains 1 vial/7.5 mL B0 calibrator and 1 vial/5.0 mL B1. These calibrators contain buffer salts, bovine serum albumin and <0.15% sodium azide; the B0 calibrator may also be used for dilution of high samples.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Cedia Carbamazepine II

Cedia Valproic Acid II

Cedia DAU 5-Drug Calibrators

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

k914857

k930734

k935792

3. Comparison with predicate:

Carbamazepine – Similarities		
Item	Device	Predicate
Indications	Measurements are used in the diagnosis and treatment of carbamazepine overdose and in monitoring levels of carbamazepine to ensure proper therapy.	Same
Storage	2 to 8°C.	Same
Technology	Quantitative, homogenous enzyme immunoassay	Same

Carbamazepine – Differences		
Item	Device	Predicate
Instrument	T60 Clinical Chemistry Analyzers	Roche Hitachi 911/912 Analyzers
Range	1.0 to 19.0 µg/ml	0.5 to ~20 µg/ml
Matrix	Serum	Serum and Plasma

Valproic Acid – Similarities		
Item	Device	Predicate
Indications	Measurements are used in the diagnosis and treatment of valproic acid overdose and in monitoring levels of valproic acid to ensure proper therapy.	Same
Technology	Quantitative, homogenous enzyme immunoassay	Same
Storage	2 to 8°C	Same
Valproic Acid – Differences		
Item	Device	Predicate
Instrument	T60 Clinical Chemistry Analyzers	Roche Hitachi 912
Range	3.0 µg/ml to 142.5 µg/ml	3.0 µg/ml to ~150 µg/ml

Valproic Acid – Similarities		
Item	Device	Predicate
Matrix	Serum	Serum or Plasma

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

CLSI - Evaluation of Precision Performance of Clinical Chemistry Devices - EP05-A2

CLSI - Evaluation of the Linearity of Quantitative Analytical Methods - EP06-A

CLSI - Method Comparison and Bias Estimation Using Patient Samples - EP09-A2

L. Test Principle:

The assays are based on the bacterial enzyme β -galactosidase which has been genetically engineered into two inactive fragments. These spontaneously reassociate to form fully active enzyme that in the assay formats, cleaves a substrate, generating a color change that can be measured spectrophotometrically.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

Carbamazepine

To evaluate precision, the within-lab and repeatability of the T60 Carbamazepine assay was calculated using the CLSI EP5-A2 method. Three commercially marketed controls (low, mid, high) were tested in duplicate during two runs per day over 21 days. The results are presented below:

Mean Concentration ($\mu\text{g/mL}$)	3.0		9.5		15.0	
	SD	CV%	SD	CV%	SD	CV%
Within-run	0.09	2.8	0.14	1.5	0.16	1.1
Between-run	0.07	2.4	-	-	0.14	0.9
Total	0.19	6.3	0.32	3.3	0.42	2.8

Valproic Acid

To evaluate precision, the within-lab and repeatability of the T60 Valproic Acid assay was calculated using the CLSI EP5-A2 method. Three commercially marketed controls (low, mid, high) were tested in duplicate during two runs per day over 21 days. The results were as follows:

Mean Concentration ($\mu\text{g/mL}$)	35.0		81.1		113.6	
	SD	CV%	SD	CV%	SD	CV%
Within-run	0.43	1.2	0.81	1.0	1.01	0.9
Between-run	0.61	1.8	1.08	1.3	1.07	0.9
Total	1.90	5.4	3.15	3.9	3.11	2.7

b. *Linearity/assay reportable range:*

Carbamazepine

The linearity study was performed using CLSI EP6-A as a guideline. Dilution series were made from 7 human sera samples with low levels of carbamazepine spiked over a measured range of range of 1.2 to 18.9 ug/mL and analyzed on a T60 analyzer. Four parallel measurements were made in random order. The study was performed with one reagent lot. Observed error was $\leq 5.0\%$. The claimed measuring range is 1.0 to 19.0 ug/dL.

Valproic Acid

The linearity study was performed using CLSI EP6-A as a guideline. Dilution series were made from 9 human sera samples with low levels of valproic acid spiked over a measured range of range of 4.3 to 141 ug/mL and analyzed on a T60 analyzer. Four parallel measurements were made in random order. The study was performed with one reagent lot. Observed error was $\leq 5.8\%$. The claimed reportable range for this assay is 3.0-142.5 $\mu\text{g/mL}$.

The package insert instructs customers to manually dilute (1:1) samples with TDM Calibrator B0 when results exceed the measuring range.

To validate the extended measurement range for carbamazepine, the sponsor used a high serum sample and made seven dilutions from the highest concentration down to normal human sera. The data support an extended range up to 37.5 $\mu\text{g/mL}$.

To validate the extended measuring range for valproic acid, the sponsor used a high serum sample and made eleven dilutions from the highest concentration down to normal human sera. The data support an extended range up to 282 $\mu\text{g/mL}$.

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

The TDM B Calibrators are purchased from another manufacturer and relabeled. The TDM B Calibrator target values are determined using assay specific methods on the T60. The target value is the median of all values obtained and traceable to USP reference materials. The product stability claims are as follows: Open vial at 2-8°C 60 days and Shelf life 2-8°C 24 months.

d. *Detection limit*

Carbamazepine

The lower limit of the assay (limit of blank - LOB) was determined by assaying 24 consecutive replicates of the analyte free (0 level) calibrator on the T60 Clinical Chemistry Analyzer. The LOB was calculated by multiplying the standard deviation by 3 and adding it to the absolute value of the mean. The limit of the blank was calculated to be 0.5 ug/mL.

The sponsor conducted a functional sensitivity study to determine the concentration at which acceptable assay precision is observed. A functional

sensitivity study was performed using serum samples and the T60 Clinical Chemistry Analyzer. The functional sensitivity was defined as the lowest concentration that can be measured with an inter-assay CV of 20%. Serum samples were run in duplicates of 4 and the mean and SD were calculated based on the observed results. The sponsor claimed that the functional sensitivity of the candidate device is 1.0 ug/mL, which is the lower limit of the measuring range.

Valproic Acid

The limit of detection (limit of blank -LOB) was determined by assaying 24 consecutive replicates of the analyte free (0 level) calibrator on the T60 Clinical Chemistry Analyzer. The LOB was calculated by multiplying the standard deviation by 3 and adding it to the absolute value of the mean. The limit of the blank was calculated to be 1.3 ug/mL.

The sponsor conducted a functional sensitivity study to determine the concentration at which acceptable assay precision is observed. A functional sensitivity study was performed using serum samples and the T60 Clinical Chemistry Analyzer. The functional sensitivity was defined as the lowest concentration that can be measured with an inter-assay CV of 20%. Serum samples were run in duplicates of 4 and the mean and SD were calculated based on the observed results. The sponsor claimed that the functional sensitivity of the candidate device is 3.0 ug/mL, the lower limit of the measuring range.

e. Analytical specificity:

Interference studies were performed using CLSI EP7-A as a guideline. Non-interference was defined as deviations less than or equal to $\pm 10\%$ of the initial value. The results are presented below:

Carbamazepine:

Bilirubin (total): No interference found up to 58 mg/dl

Hemolysate: No interference found up to 1000 mg/dl

Lipemia: No interference found up to 1000 mg/dl

The sponsor evaluated the specificity of the carbamazepine assay by testing the following compounds:

Compound	Concentration Tested ($\mu\text{g/ml}$)	Carbamazepine Concentration ($\mu\text{g/ml}$)	% Cross-reactivity
Amitryptiline	100	17.7	18.6
Carbamazepine-10,11-epoxide	250	18.1	7.4
Diazepam	250	12.0	4.8
Imipramine	200	11.3	5.6

Compound	Concentration Tested (µg/ml)	Carbamazepine Concentration (µg/ml)	% Cross-reactivity
Methsuximide	1000	10.0	1.0
Nortriptyline	50	16.4	17.2
Phenothiazine	200	16.4	8.6
Probenecid	500	10.1	2.0

The drugs listed below were spiked into normal human serum pools at the concentrations shown below. The manufacturer indicated that the drug levels evaluated were at a concentrations greater than what would be expected for a maximum daily dose. Results were compared to those of control samples without cross-reactant. The drugs and concentrations tested are shown below. Less than 1.0% cross-reactivity was observed for the following compounds:

Compound	Concentration Tested (µg/ml)
2-Phenyl-2-ethylmalonamide	1000
5-(p-Hydroxyphenyl)- phenylhydantoin	1000
Amobarbital	1000
Chlorazepate	200
Chlordiazepoxide	1000
Ethosuximide	1000
Ethotoin	1000
Glutethimide	1000
Mephentyoin	1000
p-Hydroxyphenobarbital	1000
Phenytoin	1000
Primidone	200
Promethazine	1000
Secobarbital	1000
Valproic Acid	700

Interference studies for valproic acid were performed using CLSI EP7-A as a guideline. The results are presented below:

Valproic Acid:

Bilirubin (total): No interference found up to 58 mg/dl

Hemolysate: No interference found up to 1000 mg/dl

Lipemia: No interference found up to 1000 mg/dl

The following compounds were tested for cross-reactivity in serum samples. Results were compared to those of control samples without cross-reactant.

Compound	Concentration Tested (µg/ml)	Valproic Acid Concentration (µg/ml)	% Cross-reactivity
3-Hydroxy-2-propylpentanoic acid	660	28.8	4.4
4-Hydroxy-2-propylpentanoic acid	660	28.8	4.4
5-Hydroxy-2-propylpentanoic acid	660	38.2	5.8
3-Oxo-2-propylpentanoic acid	1072	41.2	3.8
PEMA	1870	3.0	<0.16
2-Propyl-2,3-pentadienoic acid	300	42.7	14.2
2-Propyl-2-pentanoic acid	2275	22.2	1.0
2-Propyl-4-pentanoic acid	278	62.1	22.3
2-Propylglutaric acid	735	3.0	<0.4
2-Propyl-succinic acid	333	3.0	<0.9
Carbamazepine	7500	3.0	<0.04
Carbamazepine-10,11-epoxide	333	3.0	<0.9
Clonazepam	1000	3.0	<0.3
Diazepam	1000	3.0	<0.3
Phenobarbital	7500	4.5	<0.06
Phenytoin	7500	3.0	<0.04
Primidone	300	3.0	<1.0
Salicylic acid	75000	3.0	<0.004

f. Assay cut-off:

Not applicable

2. Comparison studies:

a. *Method comparison with predicate device:*

The T60 Carbamazepine assay was compared to the predicate device using CLSI Document EP9-A2 as a guideline for the method comparison study. One hundred thirty-four serum samples with Carbamazepine values ranging from 1.9 - 19.6 µg/mL as determined by the predicate device were split and run on the predicate and the proposed devices. The regression statistics are as follows:

Slope = 0.98

Intercept = 0.02 µg/mL

Correlation Coefficient = 0.993

The T60 Valproic Acid assay was compared to the predicate device using CLSI Document EP9-A2 as a guideline for the method comparison study. One hundred thirty-six serum samples with Valproic Acid values ranging from 3.2 - 143.4

µg/mL as determined by the predicate device were split and run on the predicate and the proposed devices. The regression statistics are as follows:

Slope = 0.996

Intercept = 1.4 µg/mL

Correlation Coefficient = 0.993

b. Matrix comparison:

Not applicable.

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; these are quantitative assays.

5. Expected values/Reference range:

Carbamazepine

A therapeutic range of 4 to 12 µg/mL has been reported for Carbamazepine.

See Tietz N. W. Fundamentals of Clinical Chemistry 4th edition, WB Saunders Co., Philadelphia, PA; 1996; 402-426.

Valproic Acid

A therapeutic range of 50 to 100 µg/mL has been reported for Valproic Acid.

See Tietz N. W. Fundamentals of Clinical Chemistry 4th edition, WB Saunders Co., Philadelphia, PA; 1996; 402-426.

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.