0004490819190c501V8.0

CARB2

cobas®

Carbamazepine

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
04490819 190	ONLINE TDM Carbamazepine 100 tests	System-ID 07 6905 3	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
03375790 190	Preciset TDM I calibrators CAL A-F (1 x 5 mL) Diluent (1 x 10 mL)	Codes 691-696	
04521536 190	TDM Control Set Level I (2 x 5 mL) Level II (2 x 5 mL) Level III (2 x 5 mL)	 Code 310 Code 311 Code 312	

English

System information

For **cobas c** 311/501 analyzers:

CARB2: ACN 124

For **cobas c** 502 analyzer:

CARB2: ACN 8124

Intended use

In vitro test for the quantitative determination of carbamazepine in serum and plasma on Roche/Hitachi **cobas c** systems.

Summary

Carbamazepine is an anticonvulsant drug, used in particular for the treatment of trigeminal neuralgia,[1] all forms of partial epilepsy, generalized tonic-clonic seizures, and simple and complex partial seizures.[2,3,4] The specific mechanism of carbamazepine is proposed as a depressant action on transmission through the nucleus ventralis anterior of the thalamus.[2,3] Carbamazepine, 5H-dibenz[b,f]-azepine-5-carboxamide, is an iminostilbene derivative also recognized by its common brand name, Tegretol. In the circulation, carbamazepine is approximately 70 % bound by protein.[3,4,5] The drug is metabolized to carbamazepine-10,11-epoxide, which is pharmacologically active, and then to carbamazepine-10,11-dihydroxide, both of which are excreted in urine. The plasma concentration of the epoxide metabolite ranges from 15 % to 48 % of the parent compound.[6] The epoxide has a shorter half-life (5-8 hours) than the parent compound (8-60 hours).[2,3,4] The epoxide and the 10,11-dihydroxide are excreted unaltered or after conjugation to glucuronic acid.

In combination with other clinical information, monitoring carbamazepine levels provides physicians with an effective tool to aid in adjusting dosage and achieving optimal therapeutic effect while avoiding both subtherapeutic and toxic drug levels.

Test principle

The ONLINE TDM Carbamazepine assay is a homogeneous microparticle agglutination immunoassay. It is a two-reagent system used for the detection of carbamazepine in serum. Kinetic interaction of microparticles (KIMS) will be measured using automated analyzers. In this technology biotinylated drug hapten serves as the binding partner to 1) anti-carbamazepine antibody and 2) streptavidin coated latex beads. A competitive reaction to a limited amount of specific anti-carbamazepine antibody takes place between the hapten and free carbamazepine in the serum sample. A decrease in the apparent signal is proportional to the amount of drug present in the sample.

Reagents - working solutions

R1	Carbamazepine biotinylated hapten; 2-(N-Morpholino) ethanesulfonic acid (MES) buffer, pH 6.4; preservative; surfactant
R2	Anti-carbamazepine antibody (mouse monoclonal); streptavidin coated latex microparticles: 0.08 %; N-(2-Hydroxyethyl)piperazine-N'-(2-ethanesulfonic acid) (HEPES) buffer, pH 7.5; preservative

R1 is in position B and R2 is in position C.

Precautions and warnings

For in vitro diagnostic use.
Exercise the normal precautions required for handling all laboratory reagents.

Disposal of all waste material should be in accordance with local guidelines. Safety data sheet available for professional user on request.

Reagent handling

Ready for use

Carefully invert reagent container several times prior to use to ensure that the reagent components are mixed.

Mix reagents by gentle inversion numerous times before placing on-board the analyzer.

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label
On-board in use and refrigerated on the analyzer:	30 days

Do not freeze.

Specimen collection and preparation

For specimen collection and preparation only use suitable tubes or collection containers.

Only the specimens listed below were tested and found acceptable.
Serum: Collect serum using standard sampling tubes.
Plasma: K_2- and K_3-EDTA, sodium or lithium heparin plasma.

Stability:[7]	2 days capped at 20-25 °C
	7 days capped at 4-8 °C
	4 weeks capped at -20 °C

The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.

Centrifuge samples containing precipitates before performing the assay.

Specimens should not be repeatedly frozen and thawed.

Invert thawed specimens several times prior to testing.

Usual sampling time varies dependent upon desired measurement of peak or trough values.[8] In method comparisons for serum versus sodium heparin plasma, a slope bias of up to 11 % has been observed.

Materials provided

See “Reagents – working solutions” section for reagents.

Materials required (but not provided)

See “Order information” section

General laboratory equipment

Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator’s manual for analyzer-specific assay instructions.

The performance of applications not validated by Roche is not warranted and must be defined by the user.

CARB2

Carbamazepine

cobas®

Application for serum and plasma

Deselect Automatic Rerun for these applications in the Utility menu, Application screen, Range tab.

cobas c 311 **test definition**

Assay type	2-Point End		
Reaction time /Assay points	10 / 9-52		
Wavelength (sub/main)	800 /546 nm		
Reaction direction	Increase		
Unit	µg/mL		
Reagent pipetting		Diluent (H_2O)	
R1	93 µL	–	
R2	93 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.0 µL	–	–
Decreased	2.0 µL	–	–
Increased	2.0 µL	–	–

cobas c 501/502 **test definition**

Assay type	2-Point End		
Reaction time /Assay points	10 / 15-63		
Wavelength (sub/main)	800 /546 nm		
Reaction direction	Increase		
Unit	µg/mL		
Reagent pipetting		Diluent (H_2O)	
R1	93 µL	–	
R2	93 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.0 µL	–	–
Decreased	2.0 µL	–	–
Increased	2.0 µL	–	–

Calibration

Calibrators	S1-6: Preciset TDM I calibrators
Calibration mode	RCM
Calibration frequency	6-point calibration • after **cobas c** pack change • after reagent lot change • and as required following quality control procedures

Traceability: This method has been standardized against USP reference standards. The calibrators are prepared to contain known quantities of carbamazepine in normal human serum.

Quality control

For quality control, use control materials as listed in the "Order information" section.

In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

Calculation

Roche/Hitachi **cobas c** systems automatically calculate the analyte concentration of each sample.

Conversion factor:[9] µg/mL x 4.23 = µmol/L

Limitations - interference

Criterion: Recovery within ± 10 % of initial value at carbamazepine levels of approximately 6 and 12 µg/mL (25.4 and 50.8 µmol/L).

Serum/Plasma

Icterus:[10] No significant interference up to an I index of 50 (approximate conjugated and unconjugated bilirubin concentration: 50 mg/dL or 855 µmol/L).

Hemolysis:[10] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 1000 mg/dL or 621 µmol/L).

Lipemia (Intralipid):[10] No significant interference up to an L index of 2000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

No significant interference from triglycerides up to 1000 mg/dL (11.3 mmol/L).

Rheumatoid factors: No significant interference from rheumatoid factors up to 100 IU/mL.

Total protein: No interference from total protein up to 13 g/dL.

There is the possibility that other substances and/or factors may interfere with the test and cause unreliable results.

In rare cases, falsely low results due to interference of antibodies to streptavidin can occur

Patients with renal insufficiency, such as those on hemodialysis, may exhibit carbamazepine levels in serum and/or plasma that are not consistent with clinical expectations for patients with normal renal function. This deviation with the ONLINE TDM Carbamazepine assay may be attributed to altered drug clearance in patients with compromised renal function. If results are greater than the expected range, determine if the patient has renal insufficiency. Samples with unexpectedly high results from patients with renal insufficiency should be confirmed by an alternate method.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[11]

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

ACTION REQUIRED

Special Wash Programming: The use of special wash steps is mandatory when certain test combinations are run together on Roche/Hitachi **cobas c** systems. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/Multiclean/SCCS or the NaOHD/SMS/SmpCln1+2/SCCS Method Sheets. For further instructions refer to the operator's manual. **cobas c** 502 analyzer: All special wash programming necessary for avoiding carry-over is available via the **cobas** link, manual input is not required.

Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.

Limits and ranges

Measuring range

0.35-20 µg/mL (1.5-85 µmol/L)

Manually dilute samples above the measuring range 1 + 1 with the Preciset TDM I Diluent (0 µg/mL) and reassay. Multiply the result by 2 to obtain the specimen value.

Lower limits of measurement

Lower detection limit of the test

0.35 µg/mL (1.5 µmol/L)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying two standard deviations above that of the 0 µg/mL calibrator (standard 1 + 2 SD, repeatability, n = 21).

Expected values

Investigator	**Therapeutic (µg/mL)**	**Therapeutic (µmol/L)**

CARB2

cobas®

Carbamazepine

Penry and Newmark[3]	5-12	21.2-50.8
Scheuer and Pedley[4]	8-12	33.8-50.8
Troupin et al.[12]	8-12	33.8-50.8
Strandjord and Johannessen[13]	3-12	12.7-50.8
Simonsen et al.[14]	6-10	25.4-42.3
Larkin et al.[15]	4-10	16.9-42.3
Shorvon et al.[16]	4-8	16.9-33.8
MacKichan and Kutt[17]	4-12	16.9-50.8

Equivalent diagnostic technologies have shown that in most adults receiving carbamazepine as the sole antiepileptic agent, a peak therapeutic response is achieved with levels between 8-12 µg/mL (33.8-50.8 µmol/L). Lower concentrations may provide effective therapeutic response when other anticonvulsants are used in combination with carbamazepine.[16,18]

Serum or plasma level monitoring provides an indicator for individual dosage regimen. Some patients may require levels outside these ranges for effective treatment. The ranges are therefore, provided only as a guide for interpretation along with other clinical symptoms, and are not to be taken as the sole indicator for adjustment of dosage.

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

Specific performance data[19]

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Precision was determined using human samples and controls in a modified NCCLS EP5-T2 protocol (repeatability n = 63, intermediate precision n = 63). The following results were obtained on a Roche/Hitachi **cobas c** 501 analyzer.

Serum/Plasma

Repeatability	*Mean*		*SD*		*CV*
	µg/mL	*µmol/L*	*µg/mL*	*µmol/L*	*%*
Control 1	2.7	11.4	0.1	0.4	3.5
Control 2	8.2	34.7	0.2	0.7	2.1
Control 3	13.9	58.8	0.2	0.8	1.3
HS 1	4.9	20.7	0.1	0.5	2.5
HS 2	10.3	43.6	0.2	0.8	1.7

Intermediate precision	*Mean*		*SD*		*CV*
	µg/mL	*µmol/L*	*µg/mL*	*µmol/L*	*%*
Control 1	2.7	11.4	0.1	0.4	3.4
Control 2	8.2	34.7	0.2	0.9	2.5
Control 3	13.9	58.8	0.3	1.5	2.5
HS 1	4.9	20.7	0.1	0.6	2.9
HS 2	10.3	43.6	0.2	0.9	2.1

Method comparison

Serum/plasma

Carbamazepine values for human serum and plasma samples obtained on a Roche/Hitachi **cobas c** 501 analyzer (y) were compared to those determined with the same reagent on a Roche/Hitachi 917 analyzer (x) and COBAS INTEGRA reagent on a COBAS INTEGRA 800 analyzer (x).

Roche/Hitachi 917 analyzer	Sample size (n) = 73
Passing/Bablok[20]	Linear regression
y = 0.961x + 0.025 µg/mL	y = 0.956x + 0.053 µg/mL
τ = 0.953	r = 0.995

The sample concentrations were between 0.4 and 18.5 µg/mL (1.69 and 78.3 µmol/L).

COBAS INTEGRA 800 analyzer	Sample size (n) = 53
Passing/Bablok[20]	Linear regression
y = 0.949x + 0.101 µg/mL	y = 0.939x + 0.335 µg/mL
τ = 0.917	r = 0.986

The sample concentrations were between 0.6 and 18.5 µg/mL (2.54 and 78.3 µmol/L).

Analytical specificity

The following compounds were tested for cross-reactivity.

Compound	**Concentration Tested (µg/mL)**	**% Cross-reactivity**
Carbamazepine-10,11-epoxide	29.6	14.02
10-Hydroxycarbamazepine (MHD)	100	1.19
Oxcarbazepine (Oxc)	100	0.95
Nortriptyline	50	0.48
Amitriptyline	100	0.24
Imipramine	200	0.15
Phenothiazine	200	0.11
Phenylbutazone	450	0.07
Promethazine	1000	0.04
Phenytoin	1000	0.03
Mephenytoin	1000	0.02
2-Phenyl-2-ethylmalonamide	1000	0.02
Valproic Acid	1000	0.02
Amobarbital	1000	ND
Chlordiazepoxide	30	ND
Clonazepam	12	ND
Ethosuximide	1000	ND
Ethotoin	1000	ND
Diazepam	25	ND
Glutethimide	1000	ND
Methosuximide	100	ND
p-Hydroxyphenobarbital	100	ND
5-(p-Hydroxyphenyl) -phenylhydantoin	1000	ND
Phenobarbital	1000	ND
Primidone	1000	ND
Probenecid	500	ND
Secobarbital	1000	ND

ND = Not Detected

Tests were performed on 16 drugs. No significant interference with the assay was found.

Acetaminophen	Doxycycline (Tetracycline)
Acetyl cysteine	Ibuprofen
Acetylsalicylic acid	Levodopa
Ampicillin-Na	Methyldopa + 1.5 H_2O
Ascorbic acid	Metronidazole
Ca-Dobesilate	Phenylbutazone
Cefoxitin	Rifampicin
Cyclosporine	Theophylline

cobas®

References

1 Blom S. Trigeminal neuralgia: its treatment with a new anticonvulsant drug (G-32883). Lancet 1962;839-840.

2 Eadie MJ, Tyler JH. Anticonvulsant Therapy: Pharmacological Basis and Practice. Edinburgh, Great Britain: Churchill Livingstone 1974;chap 7.

3 Penry JK, Newmark ME. The use of antiepileptic drugs. Ann Int Med 1979;90:207-218.

4 Scheuer ML, Pedley TA. The evaluation and treatment of seizures. N Engl J Med 1990;322(21):1468-1474.

5 MacKichan JJ, Duffner PK, Cohen ME. Salivary concentrations and plasma protein binding of carbamazepine and carbamazepine-10,11-epoxide in epileptic patients. Eur J Clin Pharm 1981;12:31-37.

6 Morselli PL. Carbamazepine: Absorption, distribution and excretion in complex partial seizures and their treatment. In: Penry JK, Daly DD eds. Advances in Neurology, Vol II. New York, NY: Raven Press 1975:279-293.

7 Guder W, Fonseca-Wollheim W, Heil O, et al. Maximum permissible transport and storage times for analysis of blood (serum, plasma), urine and cerebrospinal fluid. DG Klinische Chemische Mitteilungen 1995;26:205-224.

8 Jacobs DS, Kaster BL Jr, Demott WR, et al. Laboratory Test Handbook. Stowe, OH. Lexi-Compl. Mosby 1990:812.

9 Tietz NW. Fundamentals of Clinical Chemistry, 3rd ed. Pa: WB Saunders Co 1987;405:834.

10 Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-475.

11 Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240-1243.

12 Troupin A, Ojemann LM, Halpern L, et al. Carbamazepine - a double blind comparison with phenytoin. Neurology 1977;27:511-519.

13 Strandjord RE, Johannessen SI. Single-drug therapy with carbamazepine in patients with epilepsy: Serum levels and clinical effect. Epilepsia 1980;21:655-662.

14 Simonsen N, Olsen PZ, Kühl V, et al. A comparative controlled study between carbamazepine and diphenylhydantoin in psychomotor epilepsy. Epilepsia 1976;17:169-176.

15 Larkin JG, Herrick AL, McGuire GM, et al. Antiepileptic drug monitoring at the Epilepsy Clinic: A prospective evaluation. Epilepsia 1991;32:89-95.

16 Shorvon SD, Chadwick D, Galbraith AW, et al. One drug for epilepsy. Br Med J 1978;1:474-476.

17 MacKichan JJ, Kutt H. Carbamazepine: Therapeutic use and serum concentration monitoring. In: Taylor WJ, Finn AL, eds. Individualizing Drug Therapy: Practical Applications of Drug Monitoring. New York, NY: Gross Townsend & Frank Inc 1981;2:1-25.

18 Altafullah I, Talwar D, Loewenson R, et al. Factors influencing serum levels of carbamazepine and carbamazepine-10,11-epoxide in children. Epilepsy Res 1989;4(1):72-80.

19 Data on file at Roche Diagnostics.

20 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard.

CONTENT	Contents of kit
→	Volume after reconstitution or mixing

FOR US CUSTOMERS ONLY: LIMITED WARRANTY

Roche Diagnostics warrants that this product will meet the specifications stated in the labeling when used in accordance with such labeling and will be free from defects in material and workmanship until the expiration date printed on the label. THIS LIMITED WARRANTY IS IN LIEU OF ANY OTHER WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR PARTICULAR PURPOSE. IN NO EVENT SHALL ROCHE DIAGNOSTICS BE LIABLE FOR INCIDENTAL, INDIRECT, SPECIAL OR CONSEQUENTIAL DAMAGES.

COBAS, ONLINE TDM, COBAS C, PRECISET and COBAS INTEGRA are trademarks of Roche.

All other product names and trademarks are the property of their respective owners.

Significant additions or changes are indicated by a change bar in the margin.

© 2014, Roche Diagnostics

CE

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com

Distribution in USA by:
Roche Diagnostics, Indianapolis, IN
US Customer Technical Support 1-800-428-2336

Roche