Elecsys Anti-HBc II

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REF		Σ	SYSTEM
09014926160	09014926501	300	**cobas e** 402 **cobas e** 801

English

For use in the USA only

System information

Short name	ACN (application code number)
AHBC 2	10166
AHBC 2 E (for use with **cobas e** flow)	11166
AHBC 2 R (for use with **cobas e** flow)	12020

Warning

- Federal law restricts this device to sale by or on the order of a physician.
- Assay performance characteristics have not been established in patients under the age of 21, pregnant women, or in populations of immunocompromised or immunosuppressed patients.
- This assay has not been FDA licensed for the screening of blood, plasma and tissue donors.

Intended use

Immunoassay for the in vitro qualitative determination of total antibodies to hepatitis B core antigen (anti-HBc) in human serum and plasma (lithium heparin, sodium citrate, potassium EDTA) in adult patients with the symptoms of hepatitis or who may be at risk for hepatitis B virus (HBV) infection. The detection of total anti-HBc is indicative of a laboratory diagnosis for HBV infection. Further HBV serological marker testing is required to define the specific disease state. The immunoassay's performance has not been established for the monitoring of HBV disease or therapy.

The **e**lectro**c**hemi**l**uminescence **i**mmuno**a**ssay "ECLIA" is intended for use on **cobas e** immunoassay analyzers.

Summary

The hepatitis B virus consists of an outer envelope containing host-derived lipids and all S gene polypeptides, the large (L), middle (M), and small (S) surface proteins, also known as pre-S1, pre-S2 and HBsAg. The nucleocapsid contains core proteins HBcAg, a 3.2 kb, circular, partially double stranded viral DNA genome, an endogenous DNA polymerase (reverse transcriptase) enzyme, and protein kinase activity. The hepatitis core antigen comprises 183-185 amino acids.[1]

During an infection with the hepatitis B virus, antibodies to HBcAg are generally formed, which often persist for life. Anti-HBc appears shortly after the onset of infection with hepatitis B virus and can usually be detected in serum soon after the appearance of HBsAg. Anti-HBc antibodies persist both in persons who have recovered from a hepatitis B infection and in those who develop HBsAg-carrier status. Accordingly, they are an indicator of existing or past hepatitis B infection.[2]

In rare cases, an HBV infection can also run its course without the appearance of immunologically detectable anti-HBc (usually in immunosuppressed patients).[3]

Due to the long persistence of anti-HBc following a hepatitis B viral infection, screening for HBV infection may be accompanied by testing for the presence of hepatitis B core antibodies as long as those who test positive are further tested for both HBsAg and anti-HBs to differentiate infection from immunity.[4]

Determination of anti-HBc in association with other hepatitis B tests permits the diagnosis and monitoring of HBV infections. In the absence of other hepatitis B markers (HBsAg-negative persons), anti-HBc may be the only indication of an existing hepatitis B viral infection.[5]

Test principle

Competition principle. Total duration of assay: 27 minutes.

- 1st incubation: Pretreatment of 24 µL of sample with reducing agent.
- 2nd incubation: After addition of HBcAg, a complex is formed with anti-HBc antibodies in the sample.
- 3rd incubation: After addition of biotinylated antibodies and ruthenium complex[a)] -labeled antibodies specific for HBcAg, together with streptavidin-coated microparticles, the still-free binding sites on the HBc-antigens become occupied. The entire complex becomes bound to the solid phase via interaction of biotin and streptavidin.
- The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell II M. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier.
- Results are determined automatically by the software by comparing the electrochemiluminescence signal obtained from the reaction product of the sample with the signal of the cutoff value previously obtained by calibration.

a) Tris(2,2'-bipyridyl)ruthenium(II)-complex ($Ru(bpy)_3^{2+}$)

Reagents - working solutions

The **cobas e** pack (M, R1, R2) and the pretreatment reagent (RO) are labeled as AHBC 2.

M Streptavidin-coated microparticles, 1 bottle, 12.4 mL:
Streptavidin-coated microparticles 0.72 mg/mL; preservative.

R0 DTT, 1 bottle, 6.3 mL:
1,4-dithiothreitol 110 mmol/L; citrate buffer 50 mmol/L.

R1 HBcAg, 1 bottle, 15.8 mL:
HBcAg (E. coli, rDNA) > 25 ng/mL; phosphate buffer 100 mmol/L, pH 7.4; preservative.

R2 Anti-HBcAg-Ab~biotin; anti-HBcAg-Ab~$Ru(bpy)_3^{2+}$, 1 bottle, 15.8 mL:
Biotinylated monoclonal anti-HBc antibody (mouse) 700 ng/mL; monoclonal anti-HBc antibody (mouse) labeled with ruthenium complex 200 ng/mL; phosphate buffer 100 mmol/L, pH 7.4; preservative.

AHBC 2 Cal1 Negative calibrator 1, 1 bottle of 1.0 mL:
Human serum, preservative.

AHBC 2 Cal2 Positive calibrator 2, 1 bottle of 1.0 mL:
Anti-HBc (human) > 8 WHO IU/mL[b)] in human serum; preservative.

b) WHO international units

Precautions and warnings

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

Infectious or microbial waste:
Warning: handle waste as potentially biohazardous material. Dispose of waste according to accepted laboratory instructions and procedures.

Environmental hazards:
Apply all relevant local disposal regulations to determine the safe disposal.

Safety data sheet available for professional user on request.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

Warning

H317 May cause an allergic skin reaction.

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H319	Causes serious eye irritation.

Prevention:

P261	Avoid breathing mist or vapours.
P280	Wear protective gloves/ eye protection/ face protection.

Response:

P333 + P313	If skin irritation or rash occurs: Get medical advice/attention.
P337 + P313	If eye irritation persists: Get medical advice/attention.
P362 + P364	Take off contaminated clothing and wash it before reuse.

Disposal:

P501	Dispose of contents/container to an approved waste disposal plant.

Hazardous components:

- 2-methyl-2H-isothiazol-3-one hydrochloride

Product safety labeling follows EU GHS guidance.

Contact phone: 1-800-428-2336

Consider any materials of human origin as infectious and handle them using typical biosafety procedures and Universal Precautions according to the OSHA standard on Bloodborne Pathogens, 29 CFR 1910.1030.[6]

All human material should be considered potentially infectious.

The calibrators have been prepared exclusively from the blood of donors tested individually and shown to be free from HBsAg (AHBC 2 Cal1) and antibodies to HCV and HIV.

The testing methods use assays that have been approved or cleared by the FDA or that are in compliance with the legal rules of the European Union (IVDR 2017/746/EU, IVDD 98/79/EC, Annex II, List A).

The serum containing anti-HBc (AHBC 2 Cal2) was inactivated using β_2-propiolactone and UV-radiation.

However, as no inactivation or testing method can rule out the potential risk of infection with absolute certainty, the material should be handled with the same level of care as a patient specimen. In the event of exposure, the directives of the responsible health authorities should be followed.[6,7]

Avoid foam formation in all reagents and sample types (specimens, calibrators and controls).

Avoid any sample cross-contamination during preparation.

Reagent handling

The reagents (M, R0, R1, R2) in the kit are ready-for-use and are supplied in **cobas e** packs.

Calibrators:

The calibrators are supplied ready-for-use in bottles compatible with the system.

Unless the entire volume is necessary for calibration on the analyzer, transfer aliquots of the ready-for-use calibrators into empty snap-cap bottles (CalSet Vials). Attach the supplied labels to these additional bottles. Store the aliquots at 2-8 °C for later use.

Perform **only one** calibration procedure per aliquot.

All information required for correct operation is available via the **cobas** link.

Storage and stability

Store at 2-8 °C.

Do not freeze.

Store the **cobas e** pack **upright** in order to ensure complete availability of the microparticles during automatic mixing prior to use.

Stability of the **cobas e** pack:	
unopened at 2-8 °C	up to the stated expiration date
on the analyzers	16 weeks

Stability of the calibrators:	
unopened at 2-8 °C	up to the stated expiration date
after opening at 2-8 °C	16 weeks
on the analyzers at 20-25 °C	use only once

Store calibrators **upright** in order to prevent the calibrator solution from adhering to the snap-cap.

Specimen collection and preparation

Only the specimens listed below were tested and found acceptable.

Serum collected using standard sampling tubes or tubes containing separating gel.

Lithium heparin, K_2-EDTA, K_3-EDTA and sodium citrate plasma.

Test samples as soon as possible after collection. Store samples at 2-8 °C if not tested immediately.

Stable for 7 days at 20-25 °C, 14 days at 2-8 °C, 3 months at -20 °C (± 5 °C). Samples may be frozen and thawed up to 4 times.

Attention! Particularly important for the Elecsys Anti-HBc II assay: Thawed samples, samples containing precipitates, and samples for repeat measurements must be carefully centrifuged before performing the assay.

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.

Ensure the samples, calibrators and controls are at 20-25 °C prior to measurement.

Due to possible evaporation effects, samples, calibrators and controls on the analyzers should be analyzed/measured within 2 hours.

Sample stability claims were established by experimental data by the manufacturer or based on reference literature and only for the temperatures/time frames as stated in the method sheet. It is the responsibility of the individual laboratory to use all available references and/or its own studies to determine specific stability criteria for its laboratory.

Materials provided

See "Reagents – working solutions" section for reagents.

- 2 x 4 bottle labels

Materials required (but not provided)

- REF 04927931161, PreciControl Anti-HBc II, for 16 x 1.3 mL
- REF 11776576322, CalSet Vials, 2 x 56 empty snap-cap bottles
- General laboratory equipment
- **cobas e** analyzer

Additional materials for **cobas e** 402 and **cobas e** 801 analyzers:

- REF 06908799190, ProCell II M, 2 x 2 L system solution
- REF 04880293190, CleanCell M, 2 x 2 L measuring cell cleaning solution
- REF 07485409001, Reservoir Cup, 8 cups to supply ProCell II M and CleanCell M
- REF 06908853190, PreClean II M, 2 x 2 L wash solution
- REF 05694302001, Assay Tip/Assay Cup tray, 6 magazines x 6 magazine stacks x 105 assay tips and 105 assay cups, 3 wasteliners
- REF 07485425001, Liquid Flow Cleaning Cup, 2 adaptor cups to supply ISE Cleaning Solution/Elecsys SysClean for Liquid Flow Cleaning Detection Unit
- REF 07485433001, PreWash Liquid Flow Cleaning Cup, 1 adaptor cup to supply ISE Cleaning Solution/Elecsys SysClean for Liquid Flow Cleaning PreWash Unit
- REF 11298500160, ISE Cleaning Solution/Elecsys SysClean, 5 x 100 mL system cleaning solution

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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.

Resuspension of the microparticles takes place automatically prior to use.

Place the cooled (stored at 2-8 °C) **cobas e** pack on the reagent manager. Avoid foam formation. The system automatically regulates the temperature of the reagents and the opening/closing of the **cobas e** pack.

Calibrators:

Place the calibrators in the sample zone.

Read in all the information necessary for calibrating the assay.

Calibration

Traceability: This method has been standardized against the anti-HBc reference material WHO Standard (NIBSC code 95/522).

Calibration frequency: Calibration must be performed once per reagent lot using AHBC 2 Cal1, AHBC 2 Cal2 and fresh reagent (i.e. not more than 24 hours since the reagent kit was registered on the analyzer).

Renewed calibration is recommended as follows:

- after 8 weeks when using the same reagent lot
- after 28 days when using the same **cobas e** pack on the analyzer
- as required: e.g. quality control findings with PreciControl Anti-HBc II outside the defined limits

Range for electrochemiluminescence signals (counts) for the calibrators:
Negative calibrator (AHBC 2 Cal1): 100000-700000
Positive calibrator (AHBC 2 Cal2): 100-3000

Quality control

For quality control, use PreciControl Anti-HBc II.

Controls for the various concentration ranges should be run individually at least once every 24 hours when the test is in use, once per **cobas e** pack, and following each calibration.

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.

If necessary, repeat the measurement of the samples concerned.

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

The recommended quality control material is serum based. The user is responsible for providing alternate control material for plasma samples when necessary.

Results interpretation

The analyzer automatically calculates the cutoff based on the measurement of AHBC 2 Cal1 and AHBC 2 Cal2.

The result of a sample is given in the form of a cutoff (COI) index (signal sample/cutoff) with a result interpretation of:

- "non-reactive" (COI > 1.1)
- "border"[c)] (1.1 ≥ COI > 0.9)
- "reactive" (COI ≤ 0.9)

c) border = borderline

Interpretation of the results

Initial Elecsys Anti-HBc II assay result			
COI	Result	Interpretation of results	Retest procedure
> 1.1	Non-reactive[d)]	No antibodies to HBc were detected	No retest required
1.1 ≥ COI > 0.9	Border	Borderline zone (undetermined)	Retest in duplicate with the Elecsys Anti-HBc II assay
≤ 0.9	Reactive	Antibodies to HBc detected	Follow CDC recommendations for supplemental testing

d) Please note: A non-reactive anti-HBc result can indicate that the patient is either susceptible to HBV infection due to no past exposure, or is immune to HBV infection due to vaccination.

Final Elecsys Anti-HBc II assay result			
Initial result (COI)	Result after retest (COI)	Final results	Interpretation of results
> 1.1	No retest required	NON-REACTIVE[d)]	Antibodies to HBc were not detected; does not exclude the possibility of exposure to HBV
1.1 ≥ COI > 0.9	If 2 of the 3 results have a COI > 1.0	NON-REACTIVE	Antibodies to HBc were not detected; does not exclude the possibility of exposure to HBV
	If 2 of the 3 results have a COI ≤ 1.0	REACTIVE	Presumptive evidence of antibodies to HBc. Follow CDC recommendations for supplemental testing.
≤ 0.9	No retest required	REACTIVE	Presumptive evidence of antibodies to HBc. Follow CDC recommendations for supplemental testing.

Retesting of samples with an initial cutoff index 1.1 ≥ COI > 0.9 can be automatically performed (see section "**cobas e** flows").

cobas e flows

cobas e flows are procedures programmed into the system to enable a fully automated sequence of measurements and the calculation of assay combinations to perform decision algorithms.
A **cobas e** flow is available to perform a repetition of measurements in duplicate automatically for samples with an initial cutoff index 1.1 ≥ COI > 0.9.
Both subresults and the overall result message will be reported.

Cutoff determination

The cutoff value was established with in-house studies by measuring a panel of 290 samples.

A Receiver Operator Curve (ROC) analysis was used to verify the cutoff.

Validation of the cutoff was performed by external clinical studies.

Limitations

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

Current methods for the detection of antibodies to HBc may not detect all infected individuals. A non-reactive test result does not exclude the possibility of exposure to HBV.

Do not use samples and controls stabilized with azide.

Drug interference studies were performed in vitro, and may not assess the potential interferences that might be seen after the drugs are metabolized in vivo.

False positive results were observed in a limited number of patients positive for Hepatitis C Virus (HCV), Hepatitis E Virus (HEV), Human T Cell Lymphotropic Virus (HTLV) and Human Immunodeficiency Virus (HIV).

A reactive anti-HBc result does not exclude co-infection by another hepatitis virus.

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Negative anti-HBc results may occur during early infection due to delayed seroconversion.

The detection of anti-HBc antibodies indicates a present or past infection with hepatitis B virus, but does not differentiate between acute, chronic or resolved infection.

False negative results may occur due to antibody levels below the detection limit of this assay or if the patient's antibodies do not react with the antigen used in this test.

False positive results due to non-specific reactivity cannot be ruled out with the Elecsys Anti-HBc II assay.

In rare cases, interference due to extremely high titers of antibodies to streptavidin and ruthenium can occur.

Results obtained with the Elecsys Anti-HBc II immunoassay may not be used interchangeably with values obtained with different manufacturers' assay methods.

Specific performance data

All studies presented in this Method Sheet were carried out when the kits had the proprietary name of Elecsys Anti-HBc Immunoassay. The proprietary name was changed to Elecsys Anti-HBc II. There is no difference in analytical and clinical performance claims between the Elecsys Anti-HBc Immunoassay and the Elecsys Anti-HBc II. Additional sample matrix testing of K_3-EDTA was performed using Elecsys Anti-HBc II.

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

Precision

Within-laboratory precision

Repeatability and intermediate precision were determined on 1 **cobas e** 801 analyzer at 1 site using 1 lot of Elecsys reagent to test 4 serum pools and 2 levels of controls and calculated according to the CLSI (Clinical and Laboratory Standards Institute) guideline EP05-A3: 2 runs per day in duplicate each for 12 days (n = 48). The following results were obtained:

cobas e 801 analyzer					
		Repeatability		**Within-laboratory**	
Sample	Mean COI	SD COI	CV %	SD COI	CV %
HS[e)]01	1.37	0.019	1.4	0.027	1.9
HS02	0.786	0.008	1.0	0.019	2.4
HS03	0.024	0.000	1.7	0.001	3.4
HS04	2.07	0.026	1.2	0.035	1.7
PC[f)] A-HBCII1	2.12	0.030	1.4	0.038	1.8
PC A-HBCII2	0.540	0.008	1.5	0.018	3.4

e) HS = human serum

f) PC = PreciControl

Repeatability and intermediate precision were determined on 1 **cobas e** 402 analyzer at 1 site using 1 lot of Elecsys reagent to test 6 serum pools and 2 levels of controls and calculated according to the CLSI (Clinical and Laboratory Standards Institute) guideline EP05 A3: 2 runs per day in duplicate each for 12 days (n = 48). The following results were obtained:

cobas e 402 analyzer					
		Repeatability		**Within-laboratory**	
Sample	Mean COI	SD COI	CV %	SD COI	CV %
HS[e)]01	1.86	0.010	0.5	0.035	1.9
HS02	0.942	0.014	1.5	0.030	3.2
HS03	0.853	0.014	1.7	0.030	3.5
HS04	0.004	0.000	1.5	0.000	1.5
HS05	0.776	0.011	1.4	0.033	4.2
HS06	1.28	0.014	1.1	0.027	2.1
PC A-HBCII1	2.11	0.020	0.9	0.042	2.0
PC A-HBCII2	0.545	0.008	1.4	0.019	3.5

Reproducibility study

Precision was further evaluated incorporating between-run, between-day and between-site variation on the **cobas e** 801 analyzer. A reproducibility study was conducted following CLSI EP5-A3 and CLSI EP15-A3 at 3 sites incorporating a 6 member panel consisting of 1 negative, 2 high negative, 2 low positive, and 1 moderate positive human serum pools and PreciControl Anti-HBc II 1 and 2 that were assayed for 5 days, 2 runs per day, and 3 replicates per run. Data were calculated according to CLSI EP05-A3. Data from 1 reagent lot were combined to achieve SD and percent CV for repeatability, between-run, between-day, between-lot, between-site and reproducibility. The overall reproducibility (imprecision) data are summarized in the following tables:

Elecsys Anti-HBc II reproducibility on the **cobas e** 801 analyzer					
		Repeatability		**Between-run**	
Sample	Mean COI	SD COI	CV %	SD COI	CV %
HS01	1.36	0.022	1.6	0.009	0.7
HS02	0.771	0.013	1.7	0.022	2.9
HS03	0.023	0.001	2.7	0.001	2.3
HS04	2.12	0.032	1.5	0.000	0.0
HS05	1.19	0.016	1.3	0.006	0.5
HS06	0.650	0.010	1.5	0.016	2.5
PC A-HBCII1	2.14	0.032	1.5	0.011	0.5
PC A-HBCII2	0.516	0.008	1.5	0.014	2.8

Elecsys Anti-HBc II reproducibility on the **cobas e** 801 analyzer					
		Between-day		**Between-site**	
Sample	Mean COI	SD COI	CV %	SD COI	CV %
HS01	1.36	0.013	1.0	0.026	1.9
HS02	0.771	0.010	1.2	0.028	3.6
HS03	0.023	0.000	1.5	0.001	2.4
HS04	2.12	0.013	0.6	0.039	1.8
HS05	1.19	0.016	1.8	0.031	2.6
HS06	0.650	0.011	1.8	0.023	3.5
PC A-HBCII1	2.14	0.014	0.6	0.032	1.5
PC A-HBCII2	0.516	0.010	1.9	0.019	3.6

Elecsys Anti-HBc II reproducibility on the **cobas e** 801 analyzer			
		Reproducibility	
Sample	Mean COI	SD COI	CV %
HS01	1.36	0.037	2.7
HS02	0.771	0.039	5.1
HS03	0.023	0.001	4.6
HS04	2.12	0.052	2.4
HS05	1.19	0.039	3.3
HS06	0.650	0.032	4.9

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Elecsys Anti-HBc II reproducibility on the **cobas e** 801 analyzer			
		Reproducibility	
Sample	Mean COI	SD COI	CV %
PC A-HBCII1	2.14	0.048	0.027
PC A-HBCII2	0.516	0.027	5.2

Precision was further evaluated incorporating between run, between day and between site variation on the **cobas e** 402 analyzer. A reproducibility study was conducted following CLSI EP5-A3 and CLSI EP15-A3 at 3 sites incorporating a 6 member panel consisting of 2 negative, 3 low positive, and 1 moderate positive human serum pools and PreciControl Anti-HBc II 1 and 2 that were assayed for 5 days, 2 runs per day, and 3 replicates per run. Data were calculated according to CLSI EP05-A3. Data from 1 reagent lot were combined to achieve SD and percent CV for repeatability, between run, between day, between lot, between site and reproducibility. The overall reproducibility (imprecision) data are summarized in the following tables:

Elecsys Anti-HBc II reproducibility on the **cobas e** 402 analyzer					
		Repeatability		**Between-run**	
Sample	Mean COI	SD COI	CV %	SD COI	CV %
HS01	1.94	0.018	0.9	0.009	0.5
HS02	0.970	0.012	1.3	0.008	0.8
HS03	0.875	0.010	1.2	0.003	0.3
HS04	0.005	0.000	1.3	0.000	0.2
HS05	0.798	0.010	1.2	0.009	1.1
HS06	1.30	0.015	1.2	0.008	0.7
PC A-HBCII1	2.04	0.016	0.8	0.009	0.4
PC A-HBCII2	0.538	0.007	1.3	0.006	1.0

Elecsys Anti-HBc II reproducibility on the **cobas e** 402 analyzer					
		Between-day		**Between-site**	
Sample	Mean COI	SD COI	CV %	SD COI	CV %
HS01	1.94	0.007	0.3	0.028	1.5
HS02	0.970	0.000	0.0	0.020	2.0
HS03	0.875	0.007	0.8	0.020	2.3
HS04	0.005	0.000	0.0	0.000	2.2
HS05	0.798	0.008	1.0	0.018	2.3
HS06	1.30	0.000	0.0	0.017	1.3
PC A-HBCII1	2.04	0.003	0.1	0.035	1.7
PC A-HBCII2	0.538	0.008	1.5	0.015	2.7

Elecsys Anti-HBc II reproducibility on the **cobas e** 402 analyzer			
		Reproducibility	
Sample	Mean COI	SD COI	CV %
HS01	1.94	0.035	1.8
HS02	0.970	0.024	2.5
HS03	0.875	0.024	2.7
HS04	0.005	0.000	2.6
HS05	0.798	0.024	3.0
HS06	1.30	0.024	1.8
PC A-HBCII1	2.04	0.040	1.9
PC A-HBCII2	0.538	0.019	3.5

Endogenous interferences

To evaluate the effect of elevated levels of hemoglobin, bilirubin, intralipid, biotin, total protein and HAMA with the Elecsys Anti-HBc II assay on the MODULAR ANALYTICS E170 analyzer, the following samples were tested. The results are presented below:

Compound	Concentration tested
Bilirubin	≤ 428 µmol/L or ≤ 25 mg/dL
Hemoglobin	≤ 0.5 mmol/L or ≤ 0.8 g/dL
Lipemia	≤ 1000 mg/dL
Biotin	≤ 4912 nmol/L or ≤ 1200 ng/mL
Total protein	≤ 10 g/dL
HAMA	≤ 483 ng/mL

Biotin interference

This assay has no biotin interference in serum concentrations up to 1200 ng/mL. Pharmacokinetic studies have shown that serum concentrations of biotin can reach up to 355 ng/mL within the first hour after biotin ingestion for subjects consuming supplements of 20 mg biotin per day[8] and up to 1160 ng/mL for subjects after a single dose of 300 mg biotin.[9]

Drug interferences

A drug interference study was performed with 18 common therapeutic drugs, with the Elecsys Anti-HBc II assay on the MODULAR ANALYTICS E170 analyzer. Each drug was tested three-fold spiked into a negative and a low positive sample. Each drug was found to be non-interfering at the following claimed concentrations:

Compound	Concentration
Acetyl cysteine	150 mg/L
Ampicillin-Na	1000 mg/L
Ascorbic acid	300 mg/L
Ca-Dobesilate	200 mg/L
Cyclosporine	5 mg/L
Cefoxitin	2500 mg/L
Heparin	5000 U
Intralipid	10000 mg/L
Levodopa	20 mg/L
Methyldopa +1.5	20 mg/L
Metronidazole	200 mg/L
Phenylbutazone	400 mg/L
Tetracycline	50 mg/L
Acetylsalicylic acid	1000 mg/L
Rifampicin	60 mg/L
Acetaminophen	200 mg/L
Ibuprofen	500 mg/L
Theophylline	100 mg/L

Matrix effects

Studies were conducted with the Elecsys Anti-HBc II assay on the MODULAR ANALYTICS E170 analyzer to evaluate the suitability of the following five types of blood collection tubes: serum/gel separation tubes, lithium heparin plasma, K_2-EDTA plasma, K_3-EDTA plasma and sodium citrate plasma. Samples were collected into matched serum and plasma collection tubes and assayed in triplicate. The study was conducted using negative, high-negative, low-positive and positive samples for anti-HBc. The studies support the use of serum/gel separation tubes and the following plasma types: lithium heparin plasma, K_2-EDTA plasma, K_3-EDTA plasma and sodium citrate plasma. The results are shown below.

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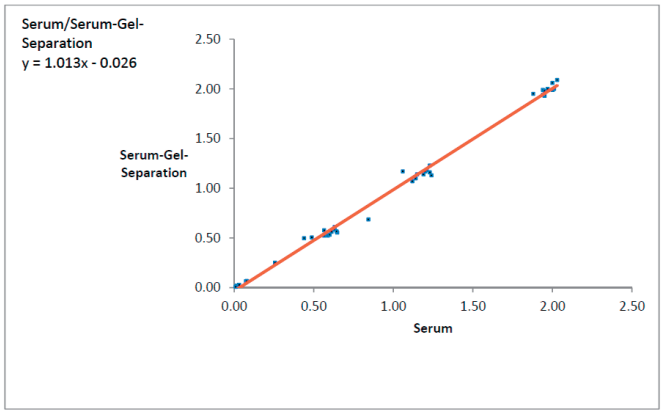

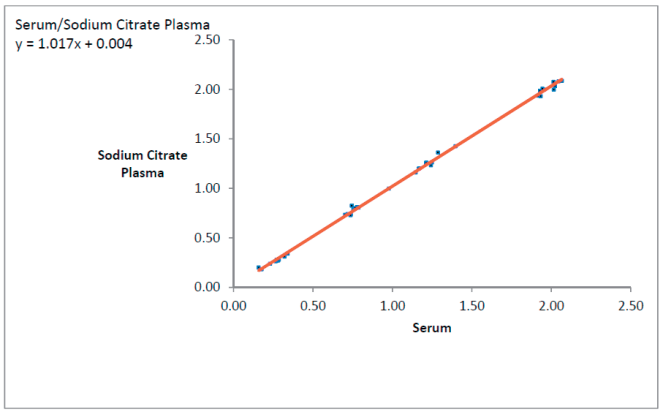

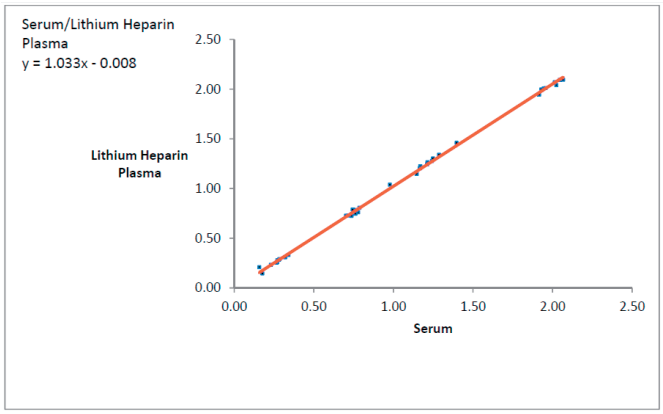

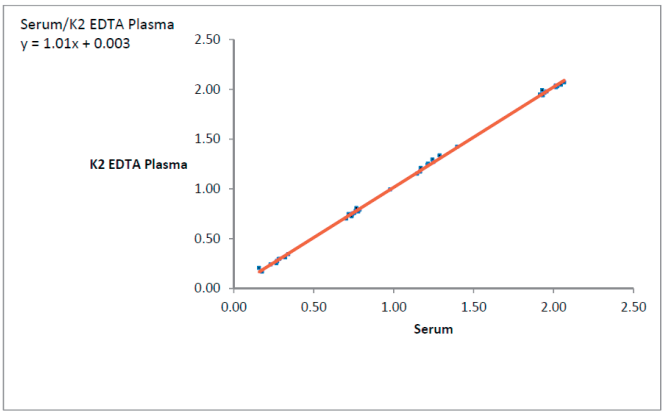

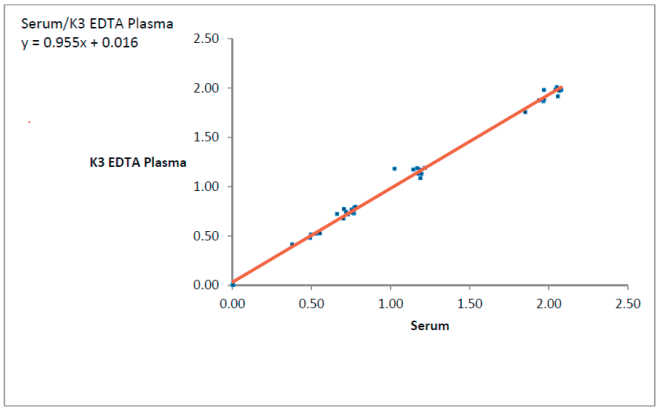

Analytical specificity

A study was conducted to evaluate the Elecsys Anti-HBc II assay on the MODULAR ANALYTICS E170 analyzer for potential cross-reactivity in specimens from individuals with medical conditions unrelated to hepatitis B infection. All specimens in the study were evaluated with the Elecsys Anti-HBc II and the reference assay. The results are summarized in the following table:

Reactivity of the Elecsys Anti-HBc II assay in individuals with medical conditions unrelated to hepatitis B infection

Category	Reference anti-HBc assay				Total
	RX[g),h)]		NR[i)]		
	Elecsys Anti-HBc II assay				
	RX	NR	RX	NR	n
Anti-nuclear antibody (ANA)	0	0	0	14	14
Cytomegalovirus (anti-CMV positive)	2	0	0	10	12
Epstein-Barr Virus (anti-EBV positive)	0	0	0	11	11
Hepatitis A Virus (anti-HAV positive)	0	0	0	9	9
HAV vaccination	0	0	0	6	6
Hepatitis C Virus (anti-HCV positive)	3	0	0	9	12
Hepatitis D Virus (anti-HDV positive)[j)]	5	0	0	0	5
Hepatitis E Virus (anti-HEV positive)	5	0	1[k)]	3	9
Human immunodeficiency virus (anti-HIV-1 positive)	5	0	1[k)]	3	9
Herpes Simplex Virus (HSV) IgG	0	0	0	9	9
Human T-Cell Lymphotropic Virus (HTLV)	2	0	1[k)]	9	12
Non-Viral Liver Disease	1	0	0	38	39
Parvovirus B_{19} infection	0	0	0	9	9
Rheumatoid Factor positive	0	0	0	11	11
Rubella	0	0	0	10	10
Syphilis (T. pallidum)	2	0	0	9	11
Toxoplasmosis IgG positive	2	0	0	6	8
Influenza vaccine recipients	0	0	0	10	10
HBV vaccination	0	0	0	7	7
E. coli infection	6	0	0	6	12
Pregnancy	0	0	0	11	11

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Reactivity of the Elecsys Anti-HBc II assay in individuals with medical conditions unrelated to hepatitis B infection					
Category	Reference anti-HBc assay				Total
	RX[g),h)]		NR[i)]		
	Elecsys Anti-HBc II assay				
	RX	NR	RX	NR	n
Varicella zoster (Anti-VZV)	1	0	0	7	8

g) RX = reactive

h) Samples that tested reactive for anti-HBc by the reference method were not further evaluated to establish the true hepatitis B infection status.

i) NR = non-reactive

j) The potential for cross-reactivity between anti-HDV reactive and anti-HBc non-reactive samples has not been established.

k) A total of three discrepant results were observed with the Elecsys Anti-HBc II assay: HEV (1/9); HTLV I/II (1/12); HIV (1/9)

Seroconversion sensitivity

Seroconversion sensitivity of the Elecsys Anti-HBc II assay tested on the **cobas e** 801 analyzer has been shown by testing 10 commercial seroconversion panels in comparison to recovery on the MODULAR ANALYTICS E170 analyzer. In 9 out of 10 panels, the Elecsys Anti-HBc II assay shows detection of seroconversion in the same bleed. The comparison of the seroconversion detection between the 2 instrument methods is summarized in the following table:

Seroconversion sensitivity of the Elecsys Anti-HBc II assay on the MODULAR ANALYTICS E170 analyzer compared to the cobas e 801 analyzer				
Seroconversion panel	Results	Number of bleeds		Discrepant results
		E170	cobas e 801	
Panel 1	Non-reactive	10	10	0
	Reactive	1	1	
Panel 2	Non-reactive	8	8	0
	Reactive	4	4	
Panel 3	Non-reactive	18	18	0
	Reactive	19	19	
Panel 4	Non-reactive	4	4	0
	Reactive	1	1	
Panel 5	Non-reactive	4	4	0
	Reactive	1	1	
Panel 6	Non-reactive	10	10	0
	Reactive	7	7	
Panel 7	Non-reactive	4	4	0
	Reactive	16	16	
Panel 8	Non-reactive	7	7	0
	Reactive	13	13	
Panel 9	Non-reactive	13	13	0
	Reactive	17	17	
Panel 10	Non-reactive	14	15	1
	Reactive	6	5	

The seroconversion study was verified on the **cobas e** 801 analyzer and **cobas e** 402 analyzer showing equivalent performance to the **cobas e** 601 analyzer.

Summary of clinical performance

Representative performance data on the MODULAR ANALYTICS E170 analyzer are given below. Results obtained in individual laboratories may differ.

A multicenter study was conducted to evaluate the ability of the Elecsys Anti-HBc II assay to detect anti-HBc antibodies in specimens from an intended use population.

Of the 1488 specimens tested in the Elecsys Anti-HBc II clinical study, 935 specimens were obtained from individuals at risk of HBV infection due to lifestyle, behavior, occupation, disease state or known exposure event; and 553 specimens were obtained from individuals with signs and symptoms of a hepatitis infection.

The 1488 specimens were collected from ten collection sites located in California (48.1 %), Florida (42.9 %), Georgia (6.05 %), and New Jersey (1.95 %). A demographic summary of the overall specimen population by age and race/ethnic group is provided in the following tables.

Demographic summary of overall specimen population by age						
	Overall		Asymptomatic		Symptomatic	
Age Group	N	%	N	%	N	%
21-30	267	17.9	159	17.0	108	19.5
31-40	306	20.6	211	22.6	95	17.2
41-50	546	36.7	352	37.7	194	35.1
51-60	313	21.0	186	19.9	127	23.0
61-70	49	3.29	23	2.46	26	4.70
71-80	7	0.470	4	0.430	3	0.540
Total	1488	100	935	100	553	100

Demographic summary of overall specimen population by race		
Race	N	%
African American/Black	710	47.7
American Indian/Alaska Native	10	0.670
Asian	6	0.400
Caucasian/White	681	45.8
Pacific Islander	4	0.270
Other	18	1.21
Unknown	59	3.97
Total	1488	100

Of the 1488 at risk subjects, 419 (28.2 %) were female and 1069 (71.8 %) were male. The mean age of the subjects was 42.6 years (age range: 21-79 years). Testing of the specimens was performed at 3 clinical testing sites located in St. Louis, MO, Ft. Lauderdale, FL and South Bend, IN.

Results by specimen classification

HBV classifications were determined based on the constellation of test results from an FDA-approved HBV marker panel. Using the reference anti-HBc assay, the specimens were assigned an HBV status based on the algorithm provided in the following table:

Serological classification by FDA-approved HBV panel						
	HBsAg	HBeAg	Anti-HBc IgM	Reference anti-HBc	Anti-HBe	Anti-HBs
Acute	(+)	(+)	(-)	(-)	(-)	(-)
Acute	(+)	(+) or (-)	eq[l)]	(+)	(+)	(-)
Acute	(+)	(+)	(+)	(+)	(-), (+), nd[m)]/ qns[n)]	(-)
Acute	(+)	(-)	(+)	(+)	(+)	(-)
Acute	(+)	qns	(+)	(+)	(+)	(-)

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Serological classification by FDA-approved HBV panel						
	HBsAg	HBeAg	Anti-HBc IgM	Reference anti-HBc	Anti-HBe	Anti-HBs
Acute	(+)	(+)	(+)	(+)	(-)	eq
Acute (Late)	(+)	(-)	(+)	(+)	(+)	(+)
Chronic	(+) > 6 mo.					
Chronic	(+) > 6 mo.	(-)	(-)	(+)	(+), eq, (+)	(-)
Chronic	(+) > 6 mo.	(+)	eq	(+)	(-)	(-)
Chronic	(+)	(+)	(-)	(+)	(-)	(-)
Chronic	(+)	(-)	(-)	(+)	(+), eq, (+)	(-)
Chronic	(+)	(+)	(+)	(+)	(-)	(+)
Early recovery	(-)	(-)	eq	(+)	(+)	(+) or eq
Early recovery	(-)	(-)	(+)	(+)	(+)	(+)
Early recovery	(-)	(-)	(-)	(+)	(+), eq, (-), qns	(-)
Recovery	(-)	(-)	(-)	(+)	(+)	(+) or eq
Recovery	(-)	(-)	(-)	(+)	eq	(+)
Recovery	(-)	(-)	(-)	(-)	(+)	(+)
Recovered or immune due to natural infection	(-)	(-)	(-)	(+)	(-)	(+) or eq
HBV vaccine response	(-)	(-)	(-)	(-)	(-)	(+)
HBV vaccine response (?)	(-)	(-)	(-)	(-)	(-)	eq
Not previously infected	(-)	(-)	(-)	(-)	(-)	(-)
Not previously infected	rr uncnf[o)]	(-)	(-)	(-)	(-)	(-)
Not interpretable	(+)	(+)	nd	(+)	(+) or (-)	(-)
Not interpretable	(+)	(-)	eq	(+)	(-)	(+)
Not interpretable	(-)	(-)	(-)	(-)	(+)	(-)
Not interpretable	(-)	(-)	(-)	(+)	qns	(+)
Not interpretable	(-)	(+)	(-)	(-)	(-)	(+) or (-)

Serological classification by FDA-approved HBV panel						
	HBsAg	HBeAg	Anti-HBc IgM	Reference anti-HBc	Anti-HBe	Anti-HBs
Not interpretable	qns	(-)	(-)	(-)	(-)	(-)

l) eq= equivocal or indeterminate or borderline

m) nd = not detected

n) qns = incomplete or unconfirmed

o) rr uncnf = repeatedly reactive: did not confirm

Results of HBV classification

Asymptomatic at risk population

The following table compares Elecsys Anti-HBc II results on the MODULAR ANALYTICS E170 analyzer with the results obtained with the reference anti-HBc assay for specimens which have been serologically classified in the asymptomatic at risk of HBV infection cohort.

Comparison of Elecsys Anti-HBc II Assay on the MODULAR ANALYTICS E170 analyzer to the Reference Assay Results by HBV Classification in the Asymptomatic At Risk Cohort

HBV classification	Reference anti-HBc assay results				Total
	+		-		
	Elecsys Anti-HBc II assay result				
	+	-	+	-	
Acute	8	0	0	1	9
Chronic	27	1	0	0	28
Early recovery	57	2	0	0	59
Recovery	133	0	1	0	134
Recovered	90	5	0	0	95
HBV vaccination	0	0	3	179	182
Not previously infected	0	0	6	416	422
Not interpretable	1	0	0	5	6
Total	316	8	10	601	935

The table below summarizes the percent agreement between the Elecsys Anti-HBc II assay and the reference assay with clinically classified samples in the asymptomatic at risk cohort. The table also provides the upper and lower 95 % exact confidence bounds.

Positive and Negative Percent Agreement in the Asymptomatic At Risk Population

HBV classification	Positive percent agreement (n/N)	95 % Exact confidence interval	Negative percent agreement (n/N)	95 % Exact confidence interval
Acute	100 (8/8)	63.1-100	100 (1/1)	2.50-100
Chronic	96.4 (27/28)	81.7-99.9	0.00 (0/0)	0.00-100
Early recovery	96.6 (57/59)	88.3-99.6	0.00 (0/0)	0.00-100
Recovery	100 (133/133)	97.3-100	0.00 (0/1)	0.00-97.5
Recovered	94.7 (90/95)	88.1-98.3	0.00 (0/0)	0.00-100
HBV vaccination	0.00 (0/0)	0.00-100	98.4 (179/182)	95.3-99.7
Not previously infected	0.00 (0/0)	0.00-100	98.6 (416/422)	96.9-99.7
Not interpretable	100 (1/1)	2.50-100	100 (5/5)	47.8-100
Total	97.5 (316/324)	95.2-98.9	98.4 (601/611)	97.0-99.2

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The positive percent agreement between the Elecsys Anti-HBc II assay results and the HBV infected status for the asymptomatic at risk for HBV infection population (n = 935) was 97.5 % (316/324) with a 95 % confidence interval of 95.2-98.9 %. The negative percent agreement between the Elecsys Anti-HBc II assay results with the not HBV infected status was 98.4 % (601/611) with a 95 % confidence interval of 97.0-99.2 %.

Symptomatic at risk population

The following table compares Elecsys Anti-HBc II results on the MODULAR ANALYTICS E170 analyzer with the results obtained with the reference anti-HBc assay for specimens which have been serologically classified in the symptomatic at risk for HBV infection cohort.

Comparison of Elecsys Anti-HBc II Assay on the MODULAR ANALYTICS E170 analyzer to the Reference Assay Results by HBV Classification in the Symptomatic At Risk Cohort

HBV classification	Reference anti-HBc assay results				Total
	+		-		
	Elecsys Anti-HBc II assay result				
	+	-	+	-	
Acute	48	0	0	0	48
Chronic	8	1	0	0	9
Early recovery	37	1	0	0	38
Recovery	68	0	0	0	68
Recovered	29	2	0	0	31
HBV vaccination	0	0	3	132	135
Not previously infected	0	0	1	218	219
Not interpretable	3	0	0	2	5
Total	193	4	4	352	553

The table below summarizes the percent agreement between the Elecsys Anti-HBc II assay on the MODULAR ANALYTICS E170 analyzer and the reference assay with clinically classified samples in the symptomatic at risk cohort. The table also provides the upper and lower 95 % exact confidence bounds.

Positive and Negative Percent Agreement in the Symptomatic At Risk Population

HBV classification	Positive percent agreement (n/N)	95 % Exact confidence interval	Negative percent agreement (n/N)	95 % Exact confidence interval
Acute	100 (48/48)	92.6-100	0.00 (0/0)	0.00-100
Chronic	88.9 (8/9)	51.8-99.7	0.00 (0/0)	0.00-100
Early recovery	97.6 (37/38)	86.2-99.9	0.00 (0/0)	0.00-100
Recovery	100 (68/68)	94.7-100	0.00 (0/0)	0.00-100
Recovered	93.6 (29/31)	78.6-99.2	0.00 (0/0)	0.00-100
HBV vaccination	0.00 (0/0)	0.00-100	97.8 (132/135)	93.6-99.5
Not previously infected	0.00 (0/0)	0.00-100	99.5 (218/219)	96.8-99.9
Not interpretable	100 (3/3)	29.2-100	100 (2/2)	15.8-100
Total	98.0 (193/197)	94.9-99.4	98.9 (352/356)	97.2-99.7

The positive percent agreement between the Elecsys Anti-HBc II assay results and the HBV infected status for the symptomatic at risk population (n = 553) was 98.0 % (193/197) with a 95 % confidence interval of 94.9-99.4 %. The negative percent agreement between the Elecsys Anti-HBc II assay results with the not HBV infected status was 99.0 % (352/356) with a 95 % confidence interval of 97.2-99.7 %.

Performance data on the cobas e 801 analyzer

All studies in this section were performed on the **cobas e** 801 analyzer.

Method comparison

A method comparison study was performed to compare the Elecsys Anti-HBc II immunoassay on the **cobas e** 801 analyzer with the MODULAR ANALYTICS E170 analyzer. Two hundred forty-one (241) human serum samples were measured on 3 different MODULAR ANALYTICS E170 analyzers and the median of the 3 MODULAR ANALYTICS E170 analyzer results was used to compare to the results obtained on the 3 different **cobas e** 801 analyzers, resulting in a total number of 723 measurements.

The negative and positive percent agreement (NPA and PPA) rates are presented in the following tables.

		Median value MODULAR ANALYTICS E170 analyzer			Total
		Reactive ≤ 0.9	Border 0.9 < x ≤ 1.1	Non-reactive > 1.1	
cobas e 801 analyzer	Reactive ≤ 0.9	390	0	0	390
	Border 0.9 < x ≤ 1.1	0	0	0	0
	Non-reactive > 1.1	0	0	333	333
Total		390	0	333	723

	Absolute	Relative	95 % CI[p)]
Negative percent agreement	333/333	100	98.90/100
Positive percent agreement	390/390	100	99.06/100

p) CI = confidence interval

The method comparison study was verified on the **cobas e** 402 analyzer showing equivalent performance to the **cobas e** 601 analyzer.

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For further information, please refer to the appropriate user guide or operator's manual for the analyzer concerned, the respective application sheets and the Method Sheets of all necessary components (if available in your country).

Elecsys Anti-HBc II

Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the Member State in which the user and/or the patient is established.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
SYSTEM	Analyzers/Instruments on which reagents can be used
REAGENT	Reagent
CALIBRATOR	Calibrator
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

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.

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