β-Hydroxybutyrate 21 FS*

Diagnostic reagent for quantitative in vitro determination of β-hydroxybutyrate in serum or plasma on photometric systems

Order Information

Cat. No.	Kit size
1 3711 99 10 930	R1 4 x 20 mL + R2 2 x 10 mL

Summary [1,2,3,4]

β-Hydroxybutyrate belongs to the group of ketone bodies and is formed during fat metabolism by reduction of acetoacetate in the liver. Ketone bodies serve as energy suppliers for various tissues (heart, kidney, and in skeletal muscles), especially in case of insulin deficiency, insulin resistance, and with low glucose concentrations. After the release of ketone bodies into the blood, they are quickly absorbed by the tissue which leads to a low concentration of ketone bodies in the blood. Metabolic acidoses due to increased β-hydroxybutyrate concentrations are related to diabetes mellitus, congenital metabolic diseases, alcoholism and fasting. Diabetic patients are at risk of developing diabetic ketoacidosis (DKA) a possible life-threatening complication. Especially diabetes patients treated by SGLT2 inhibitors are concerned, as they do not show increased glucose concentration in blood.

Method

Enzymatic determination with β-hydroxybutyrate-dehydrogenase

Principle

$$\beta\text{-Hydroxybutyrate} + NAD \xrightarrow{\beta\text{-Hydroxybutyrate-dehydrogenase}} \text{Acetoacetate} + NADH + H^+$$

The absorbance at 340 nm is proportional to the β-hydroxybutyrate concentration in the sample.

Reagents

Components and Concentrations

R1:	Buffer	pH 8.5	< 150 mmol/L
	β-Hydroxybutyrate-dehydrogenase		≥ 1 kU/L
R2:	Buffer	pH 4.3	< 70 mmol/L
	NAD		< 25 mmol/L
Standard:			1 mmol/L

Storage Instructions and Reagent Stability

The reagents and the standard are stable up to the end of the indicated month of expiry, if stored at 2 – 8°C, protected from light and contamination is avoided. Do not freeze the reagents.

Warnings and Precautions

1. Reagent 1: Warning. H319 Causes serious eye irritation. P264 Wash hands and face thoroughly after handling. P 280 Wear protective gloves/protective clothing/eye protection/face protection. P305+P351+P338 If in eyes: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. P337+ P313 If eye irritation persists: Get medical advice/attention.
2. Reagent 1 contains sodium azide (0.95 g/L) as preservative. Do not swallow! Avoid contact with skin and mucous membranes.
3. Reagent 1 contains biological material. Handle the product as potentially infectious according to universal precautions and good laboratory practice.
4. In very rare cases, samples of patients with gammopathy might give falsified results [5].
5. To avoid contamination and carryover, special care should be taken in combination with Magnesium XL FS reagent (1 4610..).
6. Refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
7. For professional use only!

Waste Management

Please refer to local legal requirements.

Reagent Preparation

The reagents and the standard are ready to use.

Materials required but not provided

NaCl solution 9 g/L
General laboratory equipment

Specimen

Serum and heparin plasma

Stability [6]:

1 month	at	20 – 25°C
1 month	at	2 – 8°C
1 month	at	–20°C

Only freeze once. Discard contaminated specimens.

Assay Procedure

Application sheets for automated systems are available on request.

Basic application parameters

Wavelength	340/700 nm (bichromatic)
Temperature	37°C
Measurement	2 Point
Sample/calibrator	12 µL
Reagent 1	160 µL
Reagent 2	40 µL
Addition Reagent 2	270 s
Absorbance A1	340 s
Absorbance A2	585 s
Calibration	Linear

ΔA = (A2 – A1) Sample/Standard

Note: For adapted procedures the volumes of sample, standard and reagents have to be calculated appropriately and the timing has to be kept exactly.

Calculation

With standard

$$\text{ß–Hydroxybutyrate [mmol/L]} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ Std.}} \times \text{Conc. Std. [mmol/L]}$$

Conversion factor

β-Hydroxybutyrate [mg/dL] x 0.096 = β-Hydroxybutyrate [mmol/L]

Calibrators and Controls

DiaSys β-Hydroxybutyrate Standard FS is recommended for calibration. β-Hydroxybutyrate Standard FS values have been made traceable to the weighing of purest β-hydroxybutyrate. Use DiaSys TruLab N and P for internal quality control. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat.-No.	Kit size
β-Hydroxybutyrate Standard FS	1 3700 99 10 030	3 x 3 mL
TruLab N	5 9000 99 10 062	20 x 5 mL
	5 9000 99 10 061	6 x 5 mL
TruLab P	5 9050 99 10 062	20 x 5 mL
	5 9050 99 10 061	6 x 5 mL

Performance Characteristics

Data evaluated on BioMajesty® JCA-BM6010/C

Measuring range	
Measuring range from 0.05 – 6.0 mmol/L β-hydroxybutyrate. When values exceed this range samples should be diluted 1 + 1 with NaCl solution (9 g/L) and the result multiplied by 2.	
Sensitivity/Limit of detection**	0.05 mmol/L

Interfering substance	**Interferences < 10% up to**	**HBUT [mmol/L]**
Acetaminophen	1.50 mmol/L	0.276
	1.50 mmol/L	4.25
Acetoacetate	5.00 mmol/L	0.267
	5.00 mmol/L	4.24
Acetylsalicylic acid	60 mg/dL	0.274
	60 mg/dL	4.27
Ascorbic acid	50 mg/dL	0.202
	50 mg/dL	2.20
Conjugated bilirubin	50 mg/dL	0.234
	50 mg/dL	2.76
Unconjugated bilirubin	50 mg/dL	0.213
	50 mg/dL	2.64
Hemoglobin	500 mg/dL	0.258
	500 mg/dL	3.04
α-Hydroxybutyrate	7.0 mmol/L	0.270
	7.0 mmol/L	1.26
Lipemia (triglycerides)	1000 mg/dL	0.256
	2000 mg/dL	2.82
NAC	1000 mg/L	0.112
	1000 mg/L	2.76
No interference by lactate and lactate dehydrogenase. For further information on interfering substances refer to Young DS [7].		

Precision			
Within run (n=20)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [mmol/L]	0.262	0.412	3.09
CV [%]	0.56	0.36	0.32
Total precision CLSI (n=80)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [mmol/L]	0.271	0.554	3.19
CV [%]	2.15	1.39	1.93

Method comparison (n=102)	
Test x	Competitor HBUT Hitachi 917
Test y	DiaSys HBUT 21 FS BioMajesty JCA-BM6010/C
Slope	1.01
Intercept	-0.014 mmol/L
Coefficient of correlation	0.999

** according to NCCLS document EP17-A2, Vol. 32, No. 8

Reference Range [1]

	[mmol/L]	[mg/dL]
Fasting	0.02 – 0.27	0.21 – 2.81

Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

Literature

1. Thomas L. Clinical Laboratory Diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft; 1998. p. 155-60.
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3. Edward C. Chao. SGLT-2 Inhibitors: A New Mechanism for Glycemic Control. Clin Diabetes 2014; 32(1): 4-11.
4. Ogawa W, Sakaguchi K. Euglycemic diabetic ketoacidosis induced by SGLT2 inhibitors: possible mechanism and contributing factors. J Diabetes Investig. 2016; 7(2):135-8.
5. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: Mechanism, detection and prevention. Clin Chem Lab Med 2007; 45(9): 1240–1243.
6. Data on file at DiaSys Diagnostic Systems GmbH.
7. Young DS. Effects of Drugs on Clinical Laboratory Tests. 5th ed. Volume 1 and 2. Washington, DC: The American Association for Clinical Chemistry Press 2000.

Manufacturer

IVD CE

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