respons®920

oneHbA1c FS*

Diagnostic reagent for quantitative in vitro determination of hemoglobin A1c (HbA1c) in whole blood on DiaSys respons®920

Order Information

Cat. No. 1 3329 99 10 922
4 twin containers (R1+R2) and 2 bottles (R3) for a total of 400 tests
Cat. No. 1 3329 99 10 927
2 twin containers (R1+R2) and 1 bottle (R3) for a total of 200 tests

Method

Particle enhanced immunoturbidimetric test
HbA1c is determined directly without measurement of total hemoglobin.

Principle

Total Hb and HbA1c in hemolyzed blood bind with the same affinity to particles in R1. The amount of binding is proportional to the relative concentration of both substances in the blood.
Mouse anti-human HbA1c monoclonal antibody (R2) binds to particle bound HbA1c. Goat anti-mouse IgG polyclonal antibody (R3) interacts with the monoclonal mouse anti-human HbA1c antibody and agglutination takes place. The measured absorbance is proportional to the HbA1c bound to particles, which in turn is proportional to the percentage of HbA1c in the sample.

Standardization

The assay is standardized according to the approved IFCC reference method [1].

NGSP and IFCC values show a linear relationship and can therefore be calculated from each other using the following equation:

HbA1c *(IFCC[b])* = (HbA1c *(NGSP[a])* – 2.15) / 0.0915

HbA1c *(NGSP[a])* = 0.0915 x HbA1c *(IFCC[b])* + 2.15

a: NGSP values in %
b: IFCC values in mmol/mol

IFCC: International Federation of Clinical Chemistry [1,2,3]
DCCT: Diabetes Control and Complications Trial [4]
NGSP: National Glycohemoglobin Standardization Program [5]

HbA1c and Average Glucose concentrations [6]

Due to a linear correlation between hemoglobin A1c and average glucose concentrations HbA1c values can be converted into estimated average glucose values by means of the following equations:
Standardization according to IFCC (calculated referring to literature reference 10):
Average glucose conc. [mg/dL] = 2.63 x HbA1c[b] + 15.01
Average glucose conc. [mmol/L] = 0.146 x HbA1c[b] + 0.829

b: HbA1c values in mmol/mol IFCC

Standardization according to NGSP:
Average glucose concentration [mg/dL] = 28.7 x HbA1c[a] – 46.7
Average glucose concentration [mmol/L] = 1.59 x HbA1c[a] – 2.59

a: HbA1c values in % NGSP

No significant differences in the regression equation were observed for variations in individuals tested, including sex, presence or absence of diabetes, type of diabetes, age, race, and ethnicity. Although this equation can be used for the majority of individuals each laboratory has to reassure itself if the regression equations mentioned are applicable for the patient group to be examined.

Reagents

Components and Concentrations

R1:	Buffer	20 mmol/L
	Latex	0.14 %
R2:	Buffer	10 mmol/L
	Mouse anti-human HbA1c monoclonal antibody	5.5 mg/dL
R3:	Buffer	10 mmol/L
	Goat anti-mouse IgG polyclonal antibody	67 mg/dL

Storage Instructions and Reagent Stability

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

Warnings and Precautions

1. Reagent 2 contains animal material. Handle the product as potentially infectious according to universal precautions and good clinical laboratory practices.
2. In very rare cases, samples of patients with gammopathy might give falsified results [13].
3. Heterophile antibodies in patient samples may cause falsified results.
4. Latex residues may necessitate cuvette wash with cleaner B (Cat. No. 1 8650 99 10 923) after use of the HbA1c test. Take care that the container with the washing solution Cleaner B is placed on the analyzer. Cuvette wash is activated by default.
5. To avoid carryover interference, please take care of efficient washing especially after use of interfering reagents. Please refer to the DiaSys respons®920 Carryover Pair Table.
6. Please 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

Reagent 2 and reagent 3 must be premixed before use. Transfer 3.5 mL R3 into the R2 cavity of the twin container. Mix very gently to avoid foaming.
Stability of premixed R2/R3: One month stored at 2 – 8°C.
With each bottle change of premixed reagent 2 and 3 a calibration must be done.

Specimen

Whole blood collected with EDTA

Please collect whole blood by standard venipuncture and fill the blood collection tube according to manufacturer specifications.

Sample preparation:

For sample preparation, DiaSys oneHbA1c Hemolyzing Solution Cat. No 1 4570 99 10 113 is required.

Sample preparation:

Hemolyzing Solution	1000 µL
Sample/Calibrator/Control	20 µL

Mix and allow to stand for 5 minutes or until complete lysis is apparent.

Stability [7]:

Whole blood	1 week	at	2 – 8°C
Hemolysate	10 hours	at	15 – 25°C
Hemolysate	10 days	at	2 – 8°C

Discard contaminated specimens.

* fluid stable

Calibrators and Controls

DiaSys TruCal HbA1c liquid is recommended for calibration. The calibrator values have been made traceable to the approved IFCC reference method. Values according to DCCT/NGSP in % have been derived from the values according to IFCC by calculation. Use DiaSys TruLab HbA1c liquid for internal quality control. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat. No.	Kit size
TruCal HbA1c liquid	1 3320 99 10 043	4 x 0.25 mL
TruLab HbA1c liquid Level 1	5 9790 99 10 074	4 x 0.25 mL
TruLab HbA1c liquid Level 2	5 9800 99 10 074	4 x 0.25 mL

Performance Characteristics

Measuring range from 30 to 150 mmol/mol HbA1c according to IFCC (4.9 to 16% according to NGSP); the assay is applicable for total hemoglobin concentrations in blood from 6.6 to 26 g/dL.	
Limit of detection (LOD/LOQ) **	30 mmol/mol HbA1c
The influence of air in opened reagent bottles may lead to reduced on board and calibration stability; therefore in R1 and R2 the use of chimneys (Cat. No. 960 637) is strongly recommended. Place chimneys into the appropriate reagent bottles. Cover reagent rotor with the RGT Bottle Locating Plate (Cat. No. 960 704).	
On board stability	4 weeks with chimney
Calibration stability	7 days with chimney

**according to CLSI Document EP17-A2, Vol. 32, No. 8

Interferences

The study on interferences was conducted according to CLSI protocol EP7-A2.

IFCC

For each interfering substance two samples with different HbA1c values have been tested; a low level sample within a HbA1c range of 20 – 40 mmol/mol and a high level sample within a HbA1c range of 60 – 100 mmol/mol.

DCCT/NGSP

For each interfering substance two samples with different HbA1c values have been tested; a low level sample within a HbA1c range of 4.0 – 5.8% and a high level sample within a HbA1c range of 7.6 – 11.3%.

The table below summarizes the results which comply for all tested levels using IFCC as well as DCCT/NGSP standardization.

Interfering substance	Interferences <7% DCCT/NGSP and < 10% IFCC
Ascorbate	up to 60 mg/dL
Bilirubin (conjugated and unconjugated)	up to 60 mg/dL
Glucose	up to 1000 mg/dL
Hemoglobin, acetylated	up to 10 mmol/L
Hemoglobin, carbamylated	up to 10 mmol/L
Lipemia (triglycerides)	up to 2000 mg/dL
N-acetylcysteine (NAC)	up to 1000 mg/L
Urea	up to 300 mg/dL
Rheumatoid factor	up to 500 IU/mL
No interference is observed by Schiff base (labile intermediates) [7]. Alcoholism and ingestion of large doses of aspirin may lead to implausible results. For further information on interfering substances refer to Young DS [11].	

Hemoglobin variants [7]:

The variants AS, AC, AD, AG, DD and elevated A2 showed no significant interferences.
The variants AE, AJ, SS, CC, SC, SE, EE, elevated F and elevated A2/F can lead to deviant HbA1c results (> 10% IFCC; > 7% DCCT/NGSP).

Precision (Values according to IFCC)			
Within run (n=20)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [mmol/mol]	32.8	55.0	83.3
Coefficient of variation [%]	2.20	1.76	1.12
Day to day (n=20)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [mmol/mol]	32.4	50.4	82.2
Coefficient of variation [%]	2.82	2.37	2.76
Total precision (CLSI) (n=80)	**Sample 1**	**Sample 2**	**Sample 3**
Mean [mmol/mol]	32.9	51.9	84.1
Coefficient of variation [%]	3.47	3.85	3.81

Method comparison (n=100) according to IFCC	
Test x	HbA1c HPLC Arkray HA-8160 V7.41
Test y	DiaSys one HbA1c FS (respons®920)
Slope	0.980
Intercept	-2.47 mmol/mol
Coefficient of correlation	0.997

Reference Range

Reference intervals should be established or verified by the laboratory based on an appropriate non-diabetic patient population.

Suggested target values for HbA1c [9]:

	mmol/mol IFCC	% NGSP
Non-diabetics	20 – 42	4 – 6
Target of therapy	< 53	< 7
Change of therapy	> 64	> 8

HbA1c cut point value for diagnosis of diabetes mellitus [12]**:**

According to a recommendation of the American Diabetes Association (ADA): ≥ 6.5% (NGSP) (48 mmol/mol (IFCC))
Patients with HbA1c values in the range of 5.7 – 6.4 % HbA1c (NGSP) or 39 – 46 mmol/mol HbA1c (IFCC) may be at high risk of developing diabetes.

Literature

1. Jeppsson OJ, Kobold U, Barr J, Finke A et al. Approved IFCC reference method for the measurement of HbA1c in human blood. Clin Chem Lab Med 2002; 40: 78–89.
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4. The Diabetes Control and Complications Trial Research Group. The effect of intensive treatment of diabetes in the development and progression of long-term complications in insulin-dependent diabetes mellitus. N Engl J Med.1993; 329: 977-86.
5. Little RR, Rohlfing CL, Wiedmeyer HM, Myers GL et al. The National Glycohemoglobin Standardization Program: A Five-Years Progress Report. Clin Chem 2001; 47: 1985-92.
6. Sacks DB. Translating Hemoglobin A1c into Average Blood Glucose: Implications for Clinical Chemistry. Clinical Chemistry 2008; 54: 1756-8.
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9. Pantheghini M, John WG on behalf of the IFCC Scientific Division. Implementation of haemoglobin A1c results traceable to the IFCC reference system: the way forward. Clin Chem Lab Med 2007; 45(8): 942-4.
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12. Sacks DB, Arnold M, Bakris GL, Bruns DE, AR Horvath et al. Guidelines and recommendations for laboratory analysis in the diagnosis and management of diabetes mellitus. Clin Chem 2011; 57(6): e1-e47.
13. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240–1243.

Manufacturer

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respons®920

ⓞⓝⓔHbA1c FS – 2 Point End

Application for hemolysate from whole blood

Test Details			Test Volumes			Reference Ranges		
Test	:	HBA1C				Auto Rerun	☐	
Report Name	:	HbA1c				Online Calibration	☐	
Unit	:	mmol/mol	Decimal Places	:	1	Cuvette Wash	☑	
Wavelength-Primary	:	660	Secondary	:	0	Total Reagents	:	2
Assay Type	:	2-Point	Curve Type	:	Exponential	Reagent R1	:	HBA1C R1
M1 Start	:	18	M1 End	:	18	Reagent R2	:	HBA1C R2
M2 Start	:	31	M2 End	:	31			
Sample Replicates	:	1	Standard Replicates	:	3	Consumables/Calibrators:		
Control Replicates	:	1	Control Interval	:	0	Blank/Level 0	:	0
Reaction Direction	:	Increasing	React. Abs. Limit	:	*	Calibrator Level 1	:	**
Prozone Limit %	:	0	Prozone Check	:	Lower	Calibrator Level 2	:	**
Linearity Limit %	:	0	Delta Abs./Min.	:	0.00	Calibrator Level 3	:	**
Technical Minimum	:	*	Technical Maximum	:	*	Calibrator Level 4	:	**
Y = aX + b a=	:	1.00	b=	:	0.00			

* HbA1c samples must not be diluted! Technical limits are automatically defined by the software via the upper and lower calibrator level.
** Enter calibrator value.

Test Details			Test Volumes	Reference Ranges
Test	:	HBA1C		
Sample Type	:	Hemolysate		

Sample Volumes

Normal	:	4.00 µL	Dilution Ratio	:	1	X
Increase	:	4.00 µL	Dilution Ratio	:	1	X
Decrease	:	4.00 µL	Dilution Ratio	:	1	X
Standard Volume	:	4.00 µL				

Reagent Volumes and Stirrer Speed

RGT-1 Volume	:	144 µL	R1 Stirrer Speed	:	High
RGT-2 Volume	:	72 µL	R2 Stirrer Speed	:	High

Sample Types

- ☑ Serum
- ☐ Urine
- ☐ CSF
- ☐ Plasma
- ☐ Whole Blood
- ☑ Other

Test Details			Test Volumes	Reference Ranges
Test	:	HBA1C		
Sample Type	:	Hemolysate		
Reference Range	:	DEFAULT		
Category	:	Male		

Reference Range

		Lower Limit (mmol/mol)	Upper Limit (mmol/mol)
Normal	:	20.00	42.00
Panic	:	0.00	0.00

Sample Types

- ☑ Serum
- ☐ Urine
- ☐ CSF
- ☐ Plasma
- ☐ Whole Blood
- ☑ Other