BECKMAN COULTER™ CE

SYNCHRON® System(s) Chemistry Information Sheet

HbA1c Hemoglobin A1c

REF 650262

For *In Vitro* Diagnostic Use

ANNUAL REVIEW

Reviewed by:	Date	Reviewed by:	Date

PRINCIPLE

INTENDED USE

The hemoglobin a1c reagent kit, when used in conjunction with SYNCHRON LX® System(s), UniCel® DxC 600/800 System(s), SYNCHRON® Systems HbA1c Calibrators and SYNCHRON® Systems Hemolyzing Reagent, is intended for the quantitative determination of hemoglobin a1c concentration as a percentage of total hemoglobin in human whole blood.

CLINICAL SIGNIFICANCE

Measurement of hemoglobin A1c is accepted as a method to measure long-term glucose control in patients with diabetes mellitus (a chronic disorder associated with disturbances in carbohydrate, fat, and protein metabolism and characterized by hyperglycemia).[1] Determination of hemoglobin A1c provides an important diagnostic tool for monitoring the efficiency of dietary control and therapy during treatment of diabetes mellitus. Long term treatment of the disease emphasizes control of blood glucose levels in preventing the acute complications of ketosis and hyperglycemia. In addition, long term complications such as retinopathy, neuropathy, and cardiovascular disease can be minimized if blood glucose levels are effectively controlled.[1,2,3]

The process of conversion from hemoglobin A to hemoglobin A1c depends on the blood glucose concentration. Since the average life of a red blood cell is 120 days, measurement of hemoglobin A1c can reflect the mean daily blood glucose concentration over the preceding two months and provides a much better indication of glycemic control than blood or urinary glucose determinations.[1,4,5,6]

Elevated levels of %HbA1c suggest the need for more aggressive treatment of glycemia. The American Diabetes Association recommends that a primary goal of therapy should be a %HbA1c < 7%, and that physicians should re-evaluate the treatment regimen in patients with %HbA1c values consistently > 8%.[7]

METHODOLOGY

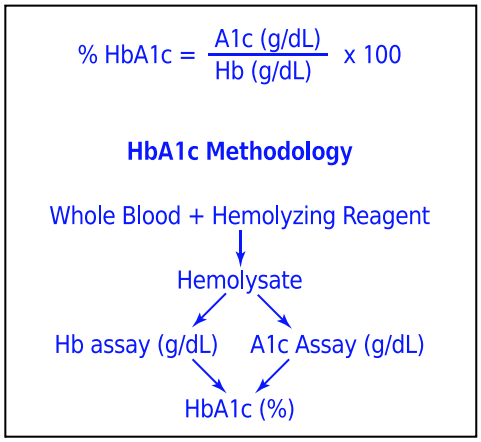
The SYNCHRON® System(s) utilizes two unique cartridges, Hb and A1c, to determine hemoglobin A1c concentration as a percentage of total hemoglobin.

Hemoglobin reagent is used to measure total hemoglobin concentration by a colorimetric method. The SYNCHRON® System(s) automatically proportions the appropriate sample and reagent volumes into the cuvette. The ratio used is one part sample to 8.6 parts reagent. The System monitors the change in absorbance at 410 nanometers. This change in

absorbance is directly proportional to the concentration of total hemoglobin in the sample and is used by the System to calculate and express total hemoglobin concentration.

A1c reagent is used to measure the hemoglobin A1c concentration by an turbidimetric immunoinhibition method. In the reaction, hemoglobin A1c antibodies combine with hemoglobin A1c from the sample to form soluble antigen-antibody complexes. Polyhaptens from the reagent then bind with the excess antibodies and the resulting agglutinated complex is measured turbidimetrically. The SYNCHRON LX System automatically proportions the appropriate sample and reagent volumes into the cuvette. The ratio used is one part sample to 31.6 parts reagent. The system monitors the change in absorbance at 340 nanometers. This change in absorbance is inversely proportional to the concentration of hemoglobin A1c in the sample and is used by the SYNCHRON LX System to calculate and express hemoglobin A1c concentration as a percentage of total hemoglobin by the following formula:

$$\% \text{ HbA1c} = \frac{\text{A1c (g/dL)}}{\text{Hb (g/dL)}} \times 100$$



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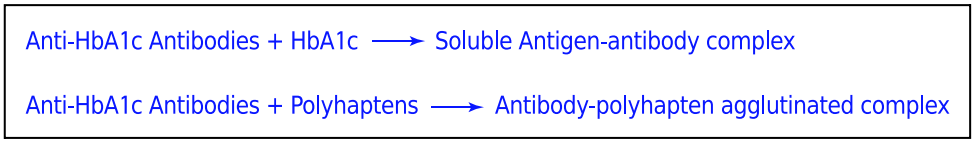
CHEMICAL REACTION SCHEME



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SPECIMEN

TYPE OF SPECIMEN

Biological fluid samples should be collected in the same manner routinely used for any laboratory test.[8] Freshly drawn blood treated with EDTA or heparin is the preferred specimen. Acceptable anticoagulants are listed in the PROCEDURAL NOTES section of this chemistry information sheet.

SPECIMEN STORAGE AND STABILITY

1. Tubes of blood are to be kept closed at all times and in a vertical, stopper-up position.[9]
2. Whole blood samples should not remain at room temperature longer than 8 hours. If assays are not completed within 8 hours, samples should be stored at +2°C to +8°C no longer than 7 days. If assays are not completed within 7 days, or the sample is to be stored beyond 7 days, samples should be frozen at -15°C to -20°C. Frozen samples are stable for 3 months and should be thawed only once. Analyte deterioration may occur in samples that are repeatedly frozen and thawed.[9]
3. Each laboratory should evaluate sample handling procedures to avoid variable results.

Additional specimen storage and stability conditions as designated by this laboratory:

SAMPLE PREPARATION

1. Bring the Hemolyzing Reagent to room temperature prior to use.
2. Pipette exactly 1000 µL Hemolyzing Reagent into a test tube. Do **NOT** pipette directly from the stock bottle.
3. Gently mix whole blood sample to ensure a uniform distribution of erythrocytes.
4. Add exactly 10 µL of whole blood sample to the test tube.
5. Rinse pipette tip in Hemolyzing Reagent by aspirating and dispensing several times.
6. Mix the hemolysate by inverting gently, avoiding the formation of foam.
7. Assay the hemolysate after hemolysis is complete, which is indicated by a color change from red to brown-green (approximately 1-2 minutes).

   Note: The hemolysate is stable for 4 hours at room temperature, or 24 hours at +2°C to +8°C.

SAMPLE VOLUME

The optimum volume, when using a 0.5 mL sample cup, is 0.3 mL of sample. For optimum primary sample tube volumes and minimum volumes, refer to the Primary Tube Sample Template for your system.

CRITERIA FOR UNACCEPTABLE SPECIMENS

Refer to the PROCEDURAL NOTES section of this chemistry information sheet for information on unacceptable specimens.

Criteria for sample rejection as designated by this laboratory:

PATIENT PREPARATION

Special instructions for patient preparation as designated by this laboratory:

SPECIMEN HANDLING

Special instructions for specimen handling as designated by this laboratory:

REAGENTS

CONTENTS

Each kit contains the following items:

Two A1c Cartridges (200 tests/cartridge)
One Hb Cartridge (400 tests/cartridge)
One Bottle Hb/A1c Calibrator Level 2 (lyophilized, 2 mL when reconstituted)
One Bottle A1c Calibrator Level 3 (lyophilized, 2 mL when reconstituted)
One Bottle A1c Calibrator Level 4 (lyophilized, 2 mL when reconstituted)
One Bottle A1c Calibrator Level 5 (lyophilized, 2 mL when reconstituted)
Two Calibrator Diskettes. Diskette #1 must be used for calibration with Synchron LX and UniCel DxC systems.
One Value Assignment Sheet

> NOTICE
>
> The components supplied in this kit are intended for use as an integral unit. Do not mix various lots of kit components.

VOLUMES PER TEST

	Hb	A1c
Sample Volume	25 µL	8 µL
Total Reagent Volume	215 µL	253 µL
Cartridge Volume		
A	215 µL	220 µL
B	– –	33 µL
C	– –	– –

REACTIVE INGREDIENTS

REAGENT CONSTITUENTS

Antibody Buffer (56 mL):	
Anti-HbA1c Antibodies (sheep)	> 0.5 mg/mL
MES (2-morpholino-ethanesulfonic acid) Buffer (pH 6.2)	0.05 mol/L
POLYHAPTEN BUFFER (12 mL):	
HbA1c Polyhapten	≥20 µg/mL
Hemoglobin Buffer (102 mL):	
Phosphate Buffer (pH 7.4)	0.02 mol/L

Also non-reactive chemicals necessary for optimal system performance.

CALIBRATOR CONSTITUENTS

Hemolysate (human and sheep)

0.9% tetradecyltrimethylammonium bromide

Also non-reactive chemicals necessary for optimal system performance.

MATERIALS NEEDED BUT NOT SUPPLIED WITH REAGENT KIT

SYNCHRON Systems Hemolyzing Reagent (for use as A1c Calibrator Level 1, and sample preparation)
At least two levels of control material

REAGENT PREPARATION

No preparation is required.

ACCEPTABLE REAGENT PERFORMANCE

The acceptability of a reagent is determined by successful calibration and by ensuring that quality control results are within your facility's acceptance criteria.

REAGENT STORAGE AND STABILITY

Hemoglobin A1c Reagent kit, when stored unopened at +2°C to +8°C, will remain stable until the expiration date printed on the kit label. Once opened, the Hemoglobin reagent cartridge is stable for 60 days at +2°C to +8°C unless the expiration date is exceeded. Once opened, the A1c reagent cartridge is stable for 30 days at +2°C to +8°C unless the expiration date is exceeded. Once opened, the SYNCHRON Systems Hemolyzing Reagent is stable until the expiration date printed on the bottle label when stored and capped at +2°C to +8°C.

Reagent storage location:

CALIBRATION

CALIBRATOR REQUIRED

Hb (Single point calibration):

SYNCHRON Hb/A1c Calibrator Level 2 (included in HbA1c reagent kit)

A1c

SYNCHRON Systems Hemolyzing Reagent (for use as A1c Calibrator Level 1)
SYNCHRON Hb/A1c Calibrator Level 2 (included in HbA1c reagent kit)
SYNCHRON A1c Calibrator Level 3 (included in HbA1c reagent kit)
SYNCHRON A1c Calibrator Level 4 (included in HbA1c reagent kit)
SYNCHRON A1c Calibrator Level 5 (included in HbA1c reagent kit)

CALIBRATOR PREPARATION

1. Carefully open calibrator bottles, avoiding loss of lyophilizate.
2. Add exactly 2000 µL of deionized water to each bottle of calibrator and replace the stopper and cap, matching each to the calibrator bottle.
3. Dissolve the contents within 30 minutes by occassional gentle swirling. Avoid the formation of foam.
4. Record calibrator reconstitution date and time on bottles.

NOTICE

Calibrators are lot-specific and should not be interchanged. Calibrators DO NOT require pretreatment with the Hemolyzing Reagent prior to assay.

CALIBRATOR STORAGE AND STABILITY

If unopened, the SYNCHRON HbA1c Calibrators should be stored at +2°C to +8°C until the expiration date printed on the calibrator bottle. Reconstituted calibrators are stable for 8 hours stored at +15°C to +25°C or 48 hours stored at +2°C to +8°C unless the expiration date is exceeded. Calibrators that are aliquoted immediately after reconstitution and stored at -15°C to -20°C are stable for 90 days. Calibrators may be frozen and thawed up to four times. Once opened, the SYNCHRON Systems Hemolyzing Reagent is stable until the expiration date printed on the bottle label when stored and capped at +2°C to +8°C.

⚠ CAUTION

Because this product is of human origin, it should be handled as though capable of transmitting infectious diseases. Each serum or plasma donor unit used in the preparation of this material was tested by United States Food and Drug Administration (FDA) approved methods and found to be negative for antibodies to HIV and HCV and nonreactive for HbsAg. Because no test method can offer complete assurance that HIV, hepatitis B virus, and hepatitis C virus or other infectious agents are absent, this material should be handled as though capable of transmitting infectious diseases. This product may also contain other human source material for which there is no approved test. The FDA recommends such samples to be handled as specified in Centers for Disease Control's Biosafety Level 2 guidelines.[10]

Calibrator storage location:

CALIBRATION INFORMATION

1. Load calibration diskette 1 only.
2. The system must have valid calibration factors in memory before controls or patient samples can be run.
3. Under typical operating conditions the Hb Reagent cartridge must be calibrated every 30 days and the A1c Reagent cartridge must be calibrated every 14 days, and also with certain parts replacement or maintenance procedures, as defined in the SYNCHRON LX *Maintenance Manual and Instrument Log*, or the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual. This assay has within-lot calibration available. Refer to the SYNCHRON LX *Operations Manual*, or the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual for information on this feature.
4. For detailed calibration instructions, refer to the SYNCHRON LX *Operations Manual*, or the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual.
5. The system will automatically perform checks on the calibration and produce data at the end of calibration. In the event of a failed calibration, the data will be printed with error codes and the system will alert the operator of the failure. For information on error codes, refer to the SYNCHRON LX *Diagnostics and Troubleshooting Manual*, or the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual.

CALIBRATOR ASSIGNED VALUES

For calibrator assigned IFCC values, see the Value Assignment Sheet provided in the Reagent Kit. Calibrator disk is supplied with each reagent kit and contain the calibrator values for that specific lot of reagent.[11,12]

CALIBRATOR SUMMARY

A one-point linear calibration scheme is used for Hb calibration. The calibration generates slope and offset that are utilized by the SYNCHRON system to convert absorbance data to Hb concentration. A five-point non-linear calibration scheme is used for A1c calibration. The calibration generates a set of calibration parameters that are utilized by the SYNCHRON system to convert absorbance data to A1c concentration.

CALIBRATOR LIMITATIONS

These calibrators should only be used in conjunction with the SYNCHRON Systems HbA1c reagent.

NOTICE
Calibrators are lot-specific and should not be interchanged.

TRACEABILITY

For value assignment information refer to the value assignment sheet in the reagent kit.

% HbA1c measurand in this calibrator is traceable to the IFCC reference Method. The traceability process is based on prEN ISO 17511.

QUALITY CONTROL

At least two levels of control material should be analyzed daily. In addition, these controls should be run with each new calibration, with each new reagent cartridge, and after specific maintenance or troubleshooting procedures as detailed in the appropriate system manual. More frequent use of controls or the use of additional controls is left to the discretion of the user based on good laboratory practices or laboratory accreditation requirements and applicable laws.

The following controls should be prepared and used in accordance with the package inserts. Discrepant quality control results should be evaluated by your facility.

Table 1.0 Quality Control Material

CONTROL NAME	SAMPLE TYPE	STORAGE

TESTING PROCEDURE(S)

1. If necessary, load the reagent onto the system.
2. After reagent load is completed, calibration may be required.
3. Program samples and controls for analysis.
4. After loading samples and controls onto the system, follow the protocols for system operations.

For detailed testing procedures, refer to the SYNCHRON LX *Operations Manual*, or the UniCel DxC 600/800 System *Instructions For Use* (IFU) manual.

CALCULATIONS

SYNCHRON® System(s) perform all calculations internally to produce the final reported result.

Calculation of the IFCC HbA1c concentration in percent is determined using the following calculation:

$$\%\ \text{HbA1c} = \frac{\text{A1c (g/dL)}}{\text{Hb (g/dL)}} \times 100$$

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Laboratories requiring certification to the National Glycohemoglobin Standardization Program (NGSP) must use the following calculation that converts the IFCC result into NGSP equivalent:

$$\%\ \text{HbA1c (NGSP)} = \frac{\text{A1c (g/dL)}}{\text{Hb (g/dL)}} \times 91.48 + 2.152$$

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Note: For automatic calculation, the equation must be entered using the special calculation function. For detailed special calculation programming, refer to Chapter 6 of the SYNCHRON® System(s) *Operations Manual*.

REPORTING RESULTS

Equivalency between the SYNCHRON LX and UniCel DxC 600/800 Systems has been established. Chemistry results between these systems are in agreement and data from representative systems may be shown.

REFERENCE INTERVALS

Each laboratory should establish its own reference intervals based upon its patient population. The NGSP reference intervals listed below were taken from literature and a study performed on SYNCHRON Systems.[13] The reference interval values were established for a population of 62 apparently healthy male and female adults from Southern California.

Table 2.0 NGSP Reference Intervals[a]

INTERVALS	SAMPLE TYPE	CONVENTIONAL UNITS
Literature	Whole Blood Hemolysate	4.0 – 6.0%
SYNCHRON	Whole Blood Hemolysate (EDTA)	4.6 – 6.2%

a Data shown was collected using SYNCHRON CX Systems. Equivalency between SYNCHRON LX Systems has been established by Deming regression analysis to SYNCHRON CX Systems.

IFCC has set 4.3% as the upper level non-diabetic guideline. Refer to IFCC website: www.ifcchba1c.com

INTERVALS	SAMPLE TYPE	CONVENTIONAL UNITS
Laboratory		

Refer to References (14,15,16) for guidelines on establishing laboratory-specific reference intervals.

Additional reporting information as designated by this laboratory:

PROCEDURAL NOTES

ANTICOAGULANT TEST RESULTS

The following anticoagulants were assessed by Deming regression analysis with a minimum of 50 paired EDTA whole blood and heparin whole blood samples. Values of EDTA (X) ranging from 4.8% HbA1c to 13.0% HbA1c were compared with the values for heparin whole blood (Y) yielding the following results.

Table 3.0 Anticoagulant Test Results[a]

ANTICOAGULANT	LEVEL OF ANTICOAGULANT TESTED	DEMING REGRESSION ANALYSIS
Lithium Heparin	14 Units/mL	Y = 0.968X + 0.12; r = 0.982
Sodium Heparin	14 Units/mL	Y = 1.006X - 0.07; r = 0.986

a Data shown was collected using SYNCHRON CX Systems. Equivalency between SYNCHRON LX Systems has been established by Deming regression analysis to SYNCHRON CX Systems.

LIMITATIONS

1. This assay is not useful in judging day-to-day glucose control and should not be used to replace daily home testing of urine and blood glucose.

2. HbA1c values are lowered in hemolytic anemia because of the shortened life of erythrocytes. Iron deficiency anemia can lead to a decreased erythrocyte mass. In either case, the average age of the erythrocytes is altered. Caution should be exercised when interpreting the HbA1c results from patients with these conditions, and when the total hemoglobin is <9 g/dL.[17,18]
3. Do not use this test for diagnosis of diabetes mellitus. Performance characteristics for this use have not been determined.
4. If unacceptable drift or imprecision is observed in Quality Control results, an optional Cuvette cleaning procedure with Cartridge Chemistry Wash Solution (CCWA, PN 657133) may be performed. The cuvette cleaning procedure is located in the As-Needed/As Required Maintenance Procedures section of the Synchron LX Maintenance Manual (CC Reagent Wash All Cuvettes) or UniCel DxC Synchron Instruction for use Manual (Wash the CC Reagent Cuvettes with CCWA).

INTERFERENCES

1. The following substances were tested for interference with this methodology:

Table 4.0 Interferences

SUBSTANCE	SOURCE	LEVEL	OBSERVED EFFECT
Bilirubin (unconjugated)	Porcine	30 mg/dL (0.3 g/L)	NSI[a]
Lipemia[b]	Human	400 mg/dL (4+) (4.0 g/L)	NSI
Rheumatoid Factor	Human	3000 IU/mL (3.0×10^6 IU/L)	NSI
Ascorbic Acid	NA[c]	50 mg/dL (0.5 g/L)	NSI

a NSI = No Significant Interference (within ±0.80% HbA1c or 10%).
b Data shown was collected using SYNCHRON CX Systems. Equivalency between SYNCHRON LX Systems has been established by Deming regression analysis to SYNCHRON CX Systems.
c NA = Not applicable.

2. Refer to References (19,20,21) for other interferences caused by drugs, disease and preanalytical variables.

SPECIFICITY

The HbA1c test shows no cross-reactivity with HbA0, HbA1a, HbA1b, acetylated hemoglobin, carbamylated hemoglobin, glycated albumin, HbA1d (an in vitro form of HbA1c formed under long storage conditions), and an acetylaldehyde hemoglobin adduct.

No effect of HbS and HbC on the assay was seen in a population of samples known to contain these variants. Likewise, no effect of HbF up to 10% (normal range <0.5%) was seen with this assay.[22] In addition, no assay interference was seen in patients with thalassemia and sickle cell disease.[23]

No effect of labile glycated hemoglobin (1000 mg/dL, 5 hours at +37°C) was seen on diabetic or non-diabetic samples.

PERFORMANCE CHARACTERISTICS

ANALYTIC RANGE

The SYNCHRON® System(s) method for the determination of percent HbA1c concentration provides the following analytical ranges:

Table 5.0 Analytical Range

ANALYTE	SAMPLE TYPE	UNITS
Hb	Whole Blood Hemolysate	6 – 24 g/dL
A1c	Whole Blood Hemolysate	0.3 to Cal 5[a]
%HbA1c	Whole Blood Hemolysate	2 – 20%

a Cal 5 Value is printed on the HbA1c2 calibrator value assignment sheet included in the kit.

If A1c concentration is below 0.3 g/dL, re-hemolyze the whole blood sample using 10 µL of sample and 500 µL of Hemolyzing Reagent and repeat both the Hb and A1c assays. No dilution factor is required for the %HbA1c result.

If A1c concentration is above the highest calibrator concentration, dilute the hemolysate 1:2 with Hemolyzing Reagent and repeat both the Hb and A1c assays. No dilution factor is required for the %HbA1c result.

REPORTABLE RANGE (AS DETERMINED ON SITE):

Table 6.0 Reportable Range

SAMPLE TYPE	CONVENTIONAL UNITS	S.I. UNITS

SENSITIVITY

Sensitivity is defined as the lowest measurable concentration which can be distinguished from zero with 95% confidence. Sensitivity for the total hemoglobin determination is 6 g/dL. Sensitivity for the A1c determination is 0.3 g/dL.

EQUIVALENCY

Equivalency was assessed by Deming regression analysis of patient samples to accepted clinical methods.

Whole Blood Hemolysate (EDTA) in the NGSP range of 3.9 to 13.1%:

Y (SYNCHRON LX Systems)	= 1.055X - 0.50
N	= 111
MEAN (SYNCHRON LX Systems)	= 7.61
MEAN (Tosoh HPLC method)	= 7.69
CORRELATION COEFFICIENT (r)	= 0.975

Refer to References (24) for guidelines on performing equivalency testing.

PRECISION

A properly operating SYNCHRON® System(s) should exhibit precision values less than or equal to the following:

Table 7.0 Precision Values

TYPE OF PRECISION	SAMPLE TYPE	1 SD	CHANGEOVER VALUE[a]	
		% HbA1c	% HbA1c	% CV
Within-run	Whole Blood Hemolysate	0.40	8.0	5.0
Total	Whole Blood Hemolysate	0.60	8.0	7.5

a When the mean of the test precision data is less than or equal to the changeover value, compare the test SD to the SD guideline given above to determine the acceptability of the precision testing. When the mean of the test precision data is greater than the changeover value, compare the test % CV to the guideline given above to determine acceptability. Changeover value = (SD guideline/CV guideline) x 100.

Comparative performance data for the SYNCHRON LX® System evaluated using the NCCLS Approved Guideline EP5-A appears in the table below.[25] Each laboratory should characterize their own instrument performance for comparison purposes.

Table 8.0 NCCLS EP5-A Precision Estimate Method (NGSP)

TYPE OF IMPRECISION	SAMPLE TYPE		No. Systems	No. Data Points[a]	Test Mean Value (% HbA1c)	EP5-A Calculated Point Estimates	
						SD	%CV
Within-run	Hemolysate	Control 1	1	80	6.05	0.17	2.84
	Hemolysate	Control 2	1	80	10.2	0.18	1.72
Total	Hemolysate	Control 1	1	80	6.05	0.24	4.0
	Hemolysate	Control 2	1	80	10.2	0.30	2.95

a The point estimate is based on the pooled data from one system, run for twenty days, two runs per day, two observations per run on an instrument operated and maintained according to the manufacturer's instructions.

NOTICE

These degrees of precision and equivalency were obtained in typical testing procedures on a SYNCHRON LX® System and are not intended to represent the performance specifications for this reagent.

ADDITIONAL INFORMATION

For more detailed information on SYNCHRON LX Systems or UniCel DxC Systems, refer to the appropriate system manual.

SHIPPING DAMAGE

If damaged product is received, notify your Beckman Coulter Clinical Support Center.

REFERENCES

1. Sperling, M. A., ed., *Physician's Guide to Insulin-Dependent (Type 1) Diabetes: Diagnosis and Treatment*, American Diabetes Association, Inc., Alexandria, VA (1988).

2. Rochman, H., "Hemoglobin A1c and Diabetes Mellitus", *Ann Clin Lab Sci.*, Vol. 10, No. 2, pp 111 115 (1980).

3. Nathan, et al., "The Effect of Intensive Treatment of Diabetes on the Development and Progression of Long Term Complications in Insulin-Dependent Diabetes Mellitus", *New England Journal of Medicine*, No. 329 pp 977 986 (1993).

4. Heinze, E., Kohne, E., Meissner, C., et al., "Hemoglobin A1c (HbA1c) in Children with Long Standing and Newly Diagnosed Diabetes Mellitus," *Acta Paediatr Scand.*, No. 68, pp 609 612 (1979).

5. Mortensen, H. B., "Glycated Hemoglobin," *Dan Med Bull.*, Vol. 32, No. 6, pp 309 328 (1985).

6. Lehmann, P., "Homogenous Immunoturbimetric Assay for Hemoglobin A1c Adaptable for Most Clinical Chemistry Analyzers: A New Concept in the Care of Diabetic Patients", AACC 45th National Meeting (1993).

7. American Diabetes Association, "Standards of Medical Care for Patients with Diabetes Mellitus", (Position statement), *Diabetes Care*, 21 (Suppl.1): S23 S31 (1998).

8. Tietz, N. W., "Specimen Collection and Processing; Sources of Biological Variation", *Textbook of Clinical Chemistry*, 2nd Edition, W. B. Saunders, Philadelphia, PA (1994).

9. National Committee for Clinical Laboratory Standards, *Procedures for the Handling and Processing of Blood Specimens*, Approved Guideline, NCCLS publication H18-A, Villanova, PA (1990).

10. CDC-NIH manual, *Biosafety in Microbiological and Biomedical Laboratories*, U.S. Government Printing Office, Washington, D.C. (1984).

11. Jeppsson, J., *Approved IFCC Reference Method for the measurement of HbA1c in Human Blood*, Clin. Chem. Lab. Med., 40(1), pp 78 89 (2002).

12. Little, R. R., *Glycated Hemoglobin Standardization - National Glycohemoglobin Standardization Program (NGSP) perspective*, Clin. Chem. Lab. Med., 41(9), pp 1191 1198 (2003).

13. Sacks, D. B., et al., "Guidelines and Recommendations for Laboratory Analysis in the Diagnosis and Management of Diabetes Mellitus", *Clinical Chemistry*, 48:3, pp 436 472 (2002).

14. National Committee for Clinical Laboratory Standards, *How to Define, Determine, and Utilize Reference Intervals in the Clinical Laboratory*, Approved Guideline, NCCLS publication C28-A, Villanova, PA (1995).

15. Tietz, N. W., *Clinical Guide to Laboratory Tests*, 3rd Edition, W. B. Saunders, Philadelphia, PA (1995).

16. Henry, J. B., *Clinical Diagnosis and Management by Laboratory Methods*, 18th Edition, W. B. Saunders Company, Philadelphia, PA (1991).

17. Greiling, H., Gressner, A. M., *Lehrbuch der Klinischen Chemie und Pathobiochemie*, 2nd Edition, Stuttgart-New York: Schattaner-Verlag; 1989:208.

18. Bessis, M., *Living Blood Cells and their Ultrastructure*, Berlin, Springer-Verlag (1973).

19. Young, D. S., *Effects of Drugs on Clinical Laboratory Tests*, 4th Edition, AACC Press, Washington, D. C. (1995).

20. Friedman, R. B., Young, D. S.,*Effects of Disease on Clinical Laboratory Tests*, 3rd Edition, AACC Press, Washington, D.C. (1997).

21. Young, D. S., *Effects of Preanalytical Variables on Clinical Laboratory Tests*, 2nd Edition, AACC Press, Washington, D. C. (1997).

22. Miedema K., *Klinishes Labor* , No. 39, pp 1029 1032 (1993).

23. Piras, G., Carluccio, A., Coe, A., Domke, I., Miravalles, E., Nanji, A., *Klinishes Labor*, No. 39, pp 1033 1037 (1993).

24. National Committee for Clinical Laboratory Standards, *Method Comparison and Bias Estimation Using Patient Samples*, Approved Guideline, NCCLS publication EP9-A, Villanova, PA (1995).

25. National Committee for Clinical Laboratory Standards, *Precision Performance of Clinical Chemistry Devices*, 2nd Edition, Approved Guideline, Vol. 19, No. 2, NCCLS publication EP5-A, Villanova, PA (1999).

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