510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY TEMPLATE

A. 510(k) Number:

k133803

B. Purpose for Submission:

New Device

C. Measurand:

Glycated Serum Protein (Fructosamine)

D. Type of Test:

Quantitative enzymatic assay

E. Applicant:

Diazyme Laboratories

F. Proprietary and Established Names:

Diazyme Glycated Serum Protein POC Test Kit

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
LCP	II	864.7470	Hematology-81

H. Intended Use:

1. Intended use(s):

   See indications for use.

2. Indication(s) for use:

The Diazyme Glycated Serum Protein POC Test Kit is intended for the quantitative determination of glycated serum proteins (GSP; fructosamine) in serum. Fructosamine is representative of blood glucose levels over the course of 2-3 weeks. The measurement of glycated serum proteins is useful for monitoring diabetic patients. For in vitro diagnostic use only.

3. Special conditions for use statement(s):

- For Prescription Use Only
- For in vitro diagnostic use
- Clinical Settings and Point-of-Care

4. Special instrument requirements:

Diazyme SMART analyzer

I. Device Description:

The Diazyme Glycated Serum Protein POC Test Kit consists of the following components (1) DRS Cuvettes pre-filled with Reagent 1 (R1), (2) DRS cap prefilled with Reagent 2 (R2) and (3) one preprogrammed Radio Frequency ID (RFID) card which contains a lot specific calibration curve. Reagent 1 is comprised of 40 prefilled DRS cuvettes and 100 mM TrisCL buffer. R2 is comprised of 40 prefilled DRS caps and enzyme/substrate reagent containing Tris.HCL buffer, enzymes, TOOS, HRP, Geneticin and stabilizers.

The SMART Analyzer (k092911) is a compact cuvette based spectrophotometer for point-of-care testing designed to analyze readings from single use reagent cuvettes. The instrument only uses the Diazyme Reagent System (DRS) cuvette and caps and performs the assay with a preprogrammed Radio Frequency ID (RFID) card. The lot specific RFID card contains reagent addition time, mixing time, reading time and calibration curve for estimating Glycated Serum Protein (GSP; fructosamine) concentration.

The Diazyme Glycated Serum Protein Controls are recommended for use with this device. The controls were previously cleared under k042193.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Diazyme Glycated Serum Protein Assay Kit

2. Predicate 510(k) number(s):

k110188

3. Comparison with predicate:

Similarities and Differences		
Item	Device Diazyme Glycated Serum Protein POC Test Kit	Predicate Diazyme Glycated Serum Protein Assay Kit k110188
Intended Use/ Indications for Use	The Diazyme Glycated Serum Protein (POC Test Kit is intended for the quantitative determination of glycated serum proteins (GSP; fructosamine) in serum. Fructosamine is representative of blood glucose levels over the course of 2-3 weeks. The measurement of glycated serum proteins is useful for monitoring diabetic patients. For in vitro diagnostic use only.	Same
Type of Test	Quantitative	Same
Methodology	Enzymatic method	Same
Sample type	Human Serum	Same
Measuring Range	61-1348 µmol/L	21-1354.0 µmol/L
Reagent	Reagent 1- 40 DRS cuvettes (prefilled) with reagent R1 • Enzyme/substrate reagent containing Tris. HCL buffer, 4-AA , Enzymes and stabilizers Reagent 2- 40 DRS caps (prefilled) • Enzyme/substrate reagent containing Tris. HCL buffer, enzymes, TOOS, HRP, Geneticin and stabilizers kit can be used on automated chemistry analyzers using validated parameters	Reagent 1-(1) bottle • Enzyme/substrate reagent containing Good's Buffer, 4-AA, enzymes and stabilizers. Reagent 2-(1) bottle • Enzyme/substrate reagent containing Good's Buffer, enzymes, TOOS, HRP, Geneticin and stabilizers
Analyzers	Kit can ONLY be used with SMART analyzers	Kit can be used on automated chemistry analyzers using validated parameters.

K. Standard/Guidance Document Referenced (if applicable):

CLSI EP5-A2: Evaluation of Precision Performance of Quantitative Measurement Methods

CLSI EP6-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach

CLSI EP7-A2: Interference Testing in Clinical Chemistry

CLSI EP9-A2: Method Comparison and Bias Estimation Using Patient Samples

CLSI EP17-A: Protocols for Determination of Limits of Detection and Limits of Quantitation

L. Test Principle:

Glycated Serum Protein (GSP; fructosamine) concentration is determined through a series of enzymatic reactions and the oxidative degradation of glycated protein fragments and uses Diazyme's specific fructosamine ™ to release hydrogen peroxide. The hydrogen peroxide released is measured by a colorimetric Trinder end-point reaction. The absorbance at 546 nm is proportional to the concentration of GSP.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

In house Precision Study: Precision studies were evaluated in accordance with the CLSI EP5-A2 guideline. Within run and total imprecision were assessed by testing a two level serum based control with the approximate concentrations of 213 and 719 µmol/L GSP (Fructosamine) respectively. An additional five serum samples with approximate GSP (Fructosamine) concentrations of 85, 225, 480,750 and 1200 µmol/L respectively were also tested. Samples were tested in 4 runs per day over 20 working days. Results are as follows:

Material	n	Mean (µmol/L)	Within-run		Total	
			SD	CV(%)	SD	CV(%)
Control 1	80	213	7.7	3.6	9.9	4.6
Control 2	80	719	22.9	3.2	28.2	3.9
Serum Level 1	80	85	5.26	6.1%	4.8	5.6
Serum Level 2	80	225	9.8	4.3	10.1	4.5

Serum Level 3	80	480	16.1	3.4	16.5	3.4
Serum Level 4	80	750	12.1	1.6	18.8	2.5
Serum Level 5	80	1200	18.6	1.5	22.7	1.8

POC Precision Study: a precision study was performed at three physician office laboratories by intended users. Each site tested 5 different patient serum samples containing different levels of GSP (Fructosamine). Each sample was analyzed in replicates of four over a five day period. The results are summarized in the tables below:

Site 1

Sample	N	Mean (µmol/L)	Within Run		Total	
			SD	%CV	SD	%CV
Sample 1	20	82	4.04	5.0	6.3	7.8
Sample 2	20	198	5.69	2.9	11.3	5.8
Sample 3	20	408	19.3	4.7	18.9	4.6
Sample 4	20	759	13.6	1.8	14.9	2.0
Sample 5	20	1138	25.7	2.3	38.4	3.4

Site 2

Sample	N	Mean (µmol/L)	Within Run		Total	
			SD	%CV	SD	%CV
Sample 6	20	92	5.6	6.1	5.6	6.1
Sample 7	20	271	15.3	5.6	13.7	5.0
Sample 8	20	451	26.7	5.9	27.7	6.2
Sample 9	20	598	37.8	6.3	34.5	5.8
Sample 10	20	1247	54.9	4.4	60.8	4.9

Site 3

Sample	N	Mean (µmol/L)	Within Run		Total	
			SD	%CV	SD	%CV
Sample 11	20	83	4.1	5.0	5.8	7.0
Sample 12	20	253	10.3	4.1	12.2	4.8
Sample 13	20	429	14.0	3.3	13.7	3.2
Sample 14	20	723	19.1	2.6	18.8	2.6
Sample 15	20	1190	21.9	1.8	19.6	1.7

b. Linearity/assay reportable range:

A linearity study was performed using 11 diluted samples with GSP (Fructosamine) concentrations evenly distributed throughout the assay range. Samples were prepared from a high serum pool spiked with human GSP (Fructosamine~1348 µmol/L) and low analyte serum GSP (Fructosamine concentration ~ 2 µmol/L) to cover the assay range. The range of samples tested was 2-1348 µmol/L. Each dilution was assayed in triplicate on the SMART analyzer. The linear regression analysis between the

expected and observed values is as follows:

y=1.007x-0.1, r=0.999

The results of the linearity study support the claimed measuring range of 61-1348 µmol/L.

c. Traceability, Stability, Expected values (controls, calibrators, or methods):

The Diazyme GSP POC Test kit is traceable to an internal standard (Master Lot) which was gravimetrically prepared from commercially available reagents. The concentrations of fructosyl propylamine were confirmed via a commercially available Fructosamine assay.

The initial lot of calibrator materials (master lot) was value assigned through a correlation study using control and patient samples with values determines using the Diazyme GSP assay (k110188) on the SMART analyzer and Randox Fructosamine assay on the Hitachi on the Hitachi 917. Based on the correlation study with this method, the fructosamine values for the Master Lot were established. All subsequent lots are value assigned against the Master Lot on the SMART analyzer.

The Diazyme POC test utilizes a Calibration Radio Frequency Identification Card (RFID) card that is programmed with a lot specific calibration curve and is supplied in each kit. RFID cards are programmed at the manufacturer site and are subject to the same quality control checks as the reagents and controls. The calibration curve is stable until the printed expiration date.

To construct the Diazyme GSP RFID card calibration curve, 2 levels of calibrators used in the predicate device (k110188) are tested with the Diazyme GSP POC Test Kit reagents on more than one SMART analyzer to obtain the rate of absorbance change. The calibrator value and the rate absorbance change are programmed into the RFID cards.

The Diazyme GSP Control Set (previously cleared under k042193) is recommended for use with this device.

d. Detection limit:

The Limit of Blank, Limit of Detection and Limit of Quantitation of the Diazyme GSP POC Test kit were determined according to CLSI EP17-A in the following manner:

To calculate the Limit of Blank (LoB) of the Diazyme GSP POC Test kit, a blank sample (water) was tested with 20 replicates daily for three days. LOB was calculated as the mean of the 57^{th} and 58^{th} highest values for the blank samples. Based upon the results, the sponsor claims a LoB=12 µmol/L.

To calculate the Limit of Detection (LoD) of the Diazyme GSP POC Test kit, five low samples (0-33µmol/L) were tested in replicates of four over three days. LoD=LoB + (1.645*SD of Low samples). Based upon the results, the sponsor claims a LoD of 29.9 µmol/L.

To calculate the Limit of Quantitation (LoQ) of the Diazyme GSP POC Test kit, 5 serum samples were prepared with GSP values at targeted concentrations (39.4 to 251.4 µmol/L were analyzed. Each serum sample was assayed on five separate runs with eight replicates per run. Based upon the results, the sponsor claims a LoQ of 61 µmol/L.

The claimed measuring range is 61-1348 µmol/L based on linearity.

The sponsor recommends that all samples >1300 µmol/L be diluted with saline and a 1:1 dilution be performed. In order to support this procedure, the sponsor provided a dilution study in which 3 serum samples were spiked and then diluted 1:1 with saline and analyzed on the SMART analyzer using the Diazyme GSP POC test. Samples ranged from 1057-1203 µmol/L and % Recovery was 97-102%

e. Analytical specificity:

The level of interference from substances normally present in serum was determined by testing two GSP serum samples (~225 µmol/L and ~550 µmol/L) spiked with various concentrations of the interferent. Test samples were then compared to a control sample (containing no interferent). The following substances produced less than 10% deviation when tested at levels equal to the concentration listed below.

Interference	Concentration
Ascorbic Acid	20 mg/dL
Bilirubin	7.5 mg/dL
Bilirubin Conjugated	5 mg/dL
Triglyceride	1000 mg/dL
Glucose	2400 mg/dL
Uric Acid	35 mg/dL
Hemoglobin*	100 mg/dL
Total Protein	12 mg/dL

*The limitations section of the labeling indicates "samples that show hemolysis should not be used for testing" with this assay.

f. Assay cut-off:

Not applicable

2. Comparison studies:

a. Method comparison with predicate device:

Internal Site Testing:

An internal method comparison study was conducted to assess the accuracy of the Diazyme Glycated Serum Protein POC test kit. A total of 54 human serum samples (were measured using the Diazyme Glycated Serum Protein POC test kit on the Diazyme SMART analyzer versus the Diazyme Glycated Serum Protein Assay Kit (k110188) on the Hitachi 917 analyzer. A total of 6 samples were altered (5 diluted and 1 spiked). Sample values ranged from 70-1269 µmol/L. The regression results are summarized below:

n	54
Slope	0.9737 (95% CI: 0.95-0.99)
Intercept	6.859(95% CI: -4.72-7.89)
Correlation coefficient (R^2)	0.997 (95% CI:0.995-0.998)
Range Tested	70-1269 µmol/L

POC Site Testing

An additional method comparison study was conducted to assess the accuracy of the Diazyme Glycated Serum Protein (POC) Test kit at the Point of Care sites. A total of 160 human serum samples were tested at three POC sites. A total of 18 samples were altered (3 spiked, 15 diluted). Each site tested between 50-60 human serum samples using the Diazyme Glycated Serum Protein POC test kit on the Diazyme SMART analyzer and compared to the results obtained using the Diazyme Glycated Serum Protein assay kit (k110188) on the Hitachi 917 analyzer. The regression results are summarized below:

Diazyme SMART GSP	Site 1	Site 2	Site 3	All sites combined
n	50	60	50	160
Slope (95% CI)	1.05 (0.99 -1.11)	1.06 (1.02-1.13)	1.02 (0.99-1.05	1.04 (1.02 – 1.07)
Intercept (95% CI)	-1.4 (15.71-16.76)	6.5 (-7.54-21.74)	-4.9 (-20.66-6.78)	-1.3 (-8.39-2.79)
Correlation Coefficient R^2 (95% CI)	0.99 (0.98-0.99)	0.99 (0.99-1.00)	0.99 (0.99-1.00)	0.99 (0.99-1.00)
Range Tested (µmol/L)	82-1330	63-1390	65-1318	63-1390

b. Matrix comparison:

Not applicable.

3. Clinical studies:

a. Clinical Sensitivity:

Not applicable

b. Clinical specificity:

Not applicable

c. Other clinical supportive data (when a. and b. are not applicable):

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

The expected normal range for non-diabetic adult Fructosamine is 100- 285 µmol/L according to literature cited by the sponsor [1,2] .

To verify the transferability of the reference interval from the predicate device, serum samples from 120 non diabetic individuals were tested using the Diazyme Glycated Serum Protein POC Test according to CLSI C28-A2 guideline. The 120 individual patient serum samples were obtained from a certified commercial source. Samples from 60 non-diabetic healthy adult males and 60 non-diabetic healthy adult females ≥18 years of age were analyzed. Results obtained confirmed the reference range cited in the literature. [1,2]

1. Wu, AHB. Tietz Clinical Guide to Laboratory Tests. Saunders 4th ed, 418:2006.
2. Schepper JD et al, Reference Values for Fructosamine Concentrations in Children's Dera: Influence of Protein Concentration, Age and Sex. Clin. Chem. 1988; 34/12:2444-2447.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.