510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY AND INSTRUMENT

I Background Information:

A 510(k) Number

K220999

B Applicant

Shijiazhuang Hipro Biotechnology Co., Ltd.

C Proprietary and Established Names

Hipro® Glycosylated Hemoglobin (HbA1c) Test System

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
PDJ	Class II	21 CFR 862.1373 - Hemoglobin A1c Test System	CH - Clinical Chemistry
LCP	Class II	21 CFR 864.7470 - Glycosylated hemoglobin assay	HE - Hematology

II Submission/Device Overview:

A Purpose for Submission:

New device

B Measurand:

Whole Blood Glycosylated Hemoglobin (HbA1c)

C Type of Test:

Quantitative Nephelometric Immunoassay

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Hipro® Glycosylated Hemoglobin (HbA1c) Test System comprised of the Hipro Glycosylated hemoglobin (HbA1c) test kit and the HP-AFS/1 automatic immunoassay analyzer is used as an aid in diagnosis of diabetes mellitus, as an aid to identify patients who may be at risk for developing diabetes mellitus, and for the monitoring of long-term blood glucose control in individuals with diabetes mellitus. It is an in vitro diagnostics reagent system intended for quantitative determination of % hemoglobin A1c (DCCT/NGSP) in venous whole blood.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

- The device should not be used to diagnose Diabetes Mellitus in patients with iron deficiency and hemolytic anemia, various hemoglobinopathies, thalassemia, hereditary spherocytosis, malignancies and severe chronic hepatic and renal disease.
- The device should not be used in pregnant patients, patients with heterozygous sickle cell trait, hemolytic diseases and recent significant or chronic blood loss.
- The device should not be used in the diagnosis of gestational diabetes.
- In cases of rapidly evolving type 1 diabetes the increase of HbA1c values might be delayed compared to the acute increase in glucose concentrations. In these conditions diabetes mellitus must be diagnosed based on plasma glucose concentration and/or the typical clinical symptoms.
- This device has significant interference with fetal hemoglobin (HbF). HbA1c results are invalid for patients with abnormal amounts of HbF including those with known Hereditary Persistence of Fetal Hemoglobin.

D Special Instrument Requirements:

The HP-AFS/1 automatic immunoassay analyzer

IV Device/System Characteristics:

A Device Description:

The Hipro® glycosylated hemoglobin (HbA1c) test system consists of the Hipro Glycosylated hemoglobin (HbA1c) test kit and the HP-AFS/1 automatic immunoassay analyzer. The Hipro Glycosylated hemoglobin (HbA1c) test kit further consists of two reagents R1 and R2. The Reagent 1 (R1) contains latex in a glycine buffer, and the Reagent 2 (R2) contains Goat anti-mouse IgG and mouse anti-human HbA1c monoclonal antibody.

B Principle of Operation:

The antigen-antibody reaction is used to directly determine the percentage of glycosylated hemoglobin (HbA1c) in total hemoglobin (Hb). The total hemoglobin and glycosylated hemoglobin in the sample have the same non-specific adsorption with latex to form a solid phase. When the specific HbA1c monoclonal antibody is added, a latex-HbA1c-mouse anti-human HbA1c monoclonal antibody complex is formed to cause agglutination due to the goat anti-mouse immunoglobulin G (IgG) antibody. The amount of agglutination is proportional to the amount of HbA1c solidified on the latex surface. The percentage of HbA1c in the sample to the total Hb can be obtained from the measured rate of change of the scattered light intensity at specific wavelength compared with the standard curve of the percentage concentration of HbA1c.

C Instrument Description Information:

1. Instrument Name:

   The HP-AFS/1 automatic immunoassay analyzer

2. Specimen Identification:

   Sample identification number can either be entered manually or scanned on barcode.

3. Specimen Sampling and Handling:

   Human venous whole blood should be collected in K2 EDTA tubes and can be stored at 2℃-8℃ for 10 days before use.

4. Calibration:

   Calibration information is provided for each new lot of the test kit through the QR code on the label of R1.

5. Quality Control:

   The sponsor recommends multi-level controls being tested in each HbA1c run using Liquidchek Diabetes Control (K123798) from Bio-Rad as the quality control material of the device. Each laboratory should establish its own control ranges and routinely use quality control materials in every HbA1c run.

V Substantial Equivalence Information:

A Predicate Device Name(s):

D-100 HbA1c, D-100 HbA1c Calibrator Pack

B Predicate 510(k) Number(s):

K151321

C Comparison with Predicate(s):

Device & Predicate Device(s):	K220999	K151321
Trade Name	Hipro Glycosylated Hemoglobin (HbA1c) Test System	D-100 HbA1c
General Device Characteristic Similarities		
Intended Use/Indications For Use	An in vitro diagnostics device intended for the quantitative determination of hemoglobin A1c in whole blood as an aid in diagnosis of diabetes, as an aid in identifying patients who may be at risk for developing diabetes mellitus, and for the monitoring of long-term blood glucose control in individuals with diabetes mellitus.	Same
Traceability	The assigned HbA1c value of the Glycosylated hemoglobin (HbA1c) test kit is certified with the National Glycohemoglobin Standardization Program (NGSP).	Same
General Device Characteristic Differences		
Methodology	Nephelometry Immunoassay Method	Ion-exchange HPLC
Measuring range	4.3-14% HbA1c	3.5- 20 % HbA1c

VI Standards/Guidance Documents Referenced:

- 21 CFR 862.1373 – Special controls for Hemoglobin A1c test system
- CLSI EP05-A3: Evaluation of Precision of Quantitative Measurement Procedures; Approved Guideline – Third Edition
- CLSI EP06-A2: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline– Second Edition
- CLSI EP07-A2: Interference Testing in Clinical Chemistry; Approved Guideline – Second Edition
- CLSI EP09-A3: Measurement Procedure Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Third Edition
- CLSI EP17-A2: Evaluation of Detection Capability for Clinical Laboratory Measurement Procedures; Approved Guideline – Second Edition

VII Performance Characteristics (if/when applicable):

A. Analytical Performance:

1. Precision/Reproducibility:

Precision studies were conducted with the Hipro Glycosylated Hemoglobin (HbA1c) Test System using four K2 EDTA venous whole blood samples containing approximately 5.2%, 6.4%, 8.0%, and 12.3% HbA1c concentrations (P1, P2, P3 and P4 respectively) and two control samples containing approximately 6% and 11% HbA1c concentrations (QC1 and QC2 respectively). Samples were analyzed on three HP-AFS/1 Automatic Immunoassay Analyzer instruments using three lots of reagents on each instrument. Each sample was analyzed in two runs per day, two replicates per run, for 20 days. The results are shown below:

Precision for Analyzer 1

Sample	Repeatability		Between-Run		Between-Day		Between-Lot		Total	
	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
P1	0.107	2.0%	0.032	0.6%	0.000	0.0%	0.000	0.0%	0.111	2.1%
P2	0.179	2.8%	0.000	0.0%	0.000	0.0%	0.029	0.5%	0.182	2.8%
P3	0.162	2.0%	0.000	0.0%	0.000	0.0%	0.020	0.2%	0.163	2.0%
P4	0.145	1.2%	0.051	0.4%	0.000	0.0%	0.017	0.1%	0.155	1.2%
QC1	0.176	2.9%	0.000	0.0%	0.000	0.0%	0.020	0.3%	0.177	2.9%
QC2	0.189	1.7%	0.000	0.0%	0.000	0.0%	0.038	0.3%	0.193	1.8%

Precision for Analyzer 2

Sample	Repeatability		Between-Run		Between-Day		Between-Lot		Total	
	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
P1	0.109	2.1%	0.000	0.0%	0.014	0.3%	0.000	0.0%	0.110	2.1%
P2	0.171	2.7%	0.000	0.0%	0.013	0.2%	0.000	0.0%	0.170	2.7%
P3	0.145	1.8%	0.000	0.0%	0.036	0.4%	0.000	0.0%	0.150	1.9%
P4	0.181	1.4%	0.026	0.2%	0.000	0.0%	0.000	0.0%	0.183	1.5%
QC1	0.177	2.9%	0.000	0.0%	0.020	0.3%	0.027	0.5%	0.180	3.0%
QC2	0.176	1.6%	0.000	0.0%	0.056	0.5%	0.000	0.0%	0.185	1.7%

Precision for Analyzer 3

Sample	Repeatability		Between-Run		Between-Day		Between-Lot		Total	
	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
P1	0.093	1.8%	0.000	0.0%	0.011	0.2%	0.000	0.0%	0.094	1.8%
P2	0.163	2.6%	0.000	0.0%	0.033	0.5%	0.000	0.0%	0.166	2.6%
P3	0.243	3.0%	0.000	0.0%	0.000	0.0%	0.022	0.3%	0.244	3.0%
P4	0.145	1.2%	0.000	0.0%	0.063	0.5%	0.050	0.4%	0.166	1.3%
QC1	0.192	3.2%	0.000	0.0%	0.000	0.0%	0.000	0.0%	0.192	3.2%
QC2	0.147	1.3%	0.000	0.0%	0.039	0.4%	0.036	0.3%	0.156	1.4%

All Analyzers Combined

Sample	Repeatability		Between-Run		Between-Day		Between-Lot		Between-Analyzer		Total	
	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
P1	0.10	2.0	0.01	0.3	0.0	0.0	0.00	0.0	0.01	0.2	0.10	2.0
P2	0.17	2.7	0.00	0.0	0.0	0.0	0.01	0.2	0.06	1.0	0.17	2.7
P3	0.19	2.3	0.00	0.0	0.00	0.0	0.02	0.2	0.03	0.4	0.19	2.4
P4	0.16	1.3	0.01	0.1	0.03	0.2	0.03	0.2	0.09	0.7	0.19	1.5
QC1	0.18	3.0	0.00	0.0	0.00	0.0	0.02	0.3	0.01	0.1	0.18	3.0
QC2	0.17	1.6	0.00	0.0	0.03	0.3	0.03	0.3	0.00	0.0	0.18	1.6

2. Linearity:

Linearity was evaluated for the Hipro Glycosylated Hemoglobin (HbA1c) Test System using a dilution series consisting of 9 levels of HbA1c prepared by mixing high and low HbA1c whole blood pools in K2 EDTA. The expected sample concentrations were 4.3%, 5.5%, 6.4%, 7.4%, 8.4%, 10.2%, 11.4%, 12.6% and 13.8% HbA1c. Three replicates were tested at each concentration. The measured values were compared to the expected values. The linear regression is the following:

$$Y=1.0225X - 0.1524,\ R^2=0.999$$

3. Analytical Specificity/Interference:

Endogenous and Exogenous Substances:
Interference studies were performed to assess the impact of endogenous and exogenous substances on the performance of the Hipro Glycosylated Hemoglobin (HbA1c) Test System. Pooled whole blood at two HbA1c levels (~6.6 and ~8.7% HbA1c) in K2 EDTA spiked with potential interferent was analyzed and compared to the same whole blood pool without interferent (control). The highest concentrations at which no significant interference (a difference of no more than ±7% from the control sample as defined by the sponsor) was observed are summarized below:

Interferent	Highest Concentration with No Significant Interference (mg/dL)
Lipemia	600
Unconjugated Bilirubin	60
Conjugated Bilirubin	60
Glucose	9,000
Rheumatoid Factor	100 IU/mL
Total Protein	15,000
Acetaminophen	20
Acetylsalicylic acid	100
Ampicillin-Na	100
Ascorbic acid	30
Cefoxitin	250
Cyclosporin	1.66
Doxycycline	5
Heparin	5000 U/L
Ibuprofen	50
Levodopa	2
Methyldopa	2
Metronidazole	20
N-Acetylcysteine	166
Phenylbutazone	40
Rifampicin	6
Theophylline	10

Cross Reactivity with Hemoglobin Derivatives:
Potential interference from HbA0, Labile HbA1c, Carbamylated Hb, Acetylated Hb, Glycated Albumin, and HbA1a+b were evaluated. Two HbA1c concentrations of pooled whole blood (~6.6% and~ 8.7%) were spiked with the prepared potential interferent. The %HbA1c values were compared to the same control sample with no potential interferent present. The highest concentrations at which no significant interference (a difference of no more than ±7% from the control sample as defined by the sponsor) was observed are summarize below:

Hemoglobin Derivatives	Highest Concentration Level Tested with No Significant Interference
Hb A0	150 g/dL
HbA1a+b	1 g/dL
Labile HbA1c	1 g/dL
Carbamylated Hb	1.5 g/dL
Acetylated Hb	1.5 g/dL
Glycated Albumin	10 g/dL

Hemoglobin Variant Interference:
A hemoglobin variant study was performed using K2 EDTA venous whole blood samples known to contain hemoglobin variants HbS, HbC, HbE, HbD, HbA2, and HbF. The samples containing the hemoglobin variants were tested using the Hipro Glycosylated Hemoglobin (HbA1c) Test System and using the comparator method (Bio-Rad VARIANT II TURBO Hemoglobin Testing System), which has been demonstrated to be free from interference by these hemoglobin variants. The following table summarizes the tested samples:

Hemoglobin Variant	Number of Samples	Variant Concentration Range (%)	Range of %HbA1c Concentration
HbS	20	16.8%-76.9%	5.1-9.4
HbC	20	25%-66.6%	4.7-9.7
HbD	20	12.9%-41.1%	5.2-10.2
HbE	20	13.8%-30.5%	5.1-12.2
HbA2	20	3.1%-6.4%	4.7-9.7
HbF	20	1.3%-40.5%	4.4-8.4

Hemoglobin Variant Results Summary

Hemoglobin Variants	Relative Bias Observed Relative to the Comparator Method			
	At ~6.5 % HbA1c		At ~9.0% HbA1c	
	Relative bias, %	Range of relative bias, %	Relative bias, %	Range of relative bias, %
HbS	1.69	-3.08~6.15	1.20	1.08~1.35
HbC	-0.17	-3.77~3.77	2.27	-1.15~4.11
HbD	1.67	-1.79~5.08	2.10	1.16~3.03
HbE	1.58	-1.67~5.36	1.38	0.00~4.17
HbA2	1.57	-1.79~5.00	-0.51	-1.02~0.00
HbF	Specimens containing HbF (>8%) may yield inaccurate HbA1c with higher variability			

The results show that there is no significant interference for HbS, HbC, HbE, HbD, and HbA2 at the concentrations stated in the table above.

The sponsor has included the following prominent boxed warning in the labeling: **This device has significant negative interference with fetal hemoglobin (HbF). HbA1c results are invalid for patients with abnormal amounts of HbF including those with known Hereditary Persistence of Fetal Hemoglobin.**

4. Assay Reportable Range:

The reportable range for this device is 4.3-14.0% HbA1c.

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

The assigned HbA1c values of the Hipro glycosylated hemoglobin (HbA1c) test kit are certified by the National Glycohemoglobin Standardization Program (NGSP). The NGSP certification expires in one year. See the NGSP website for current certification at http://www.ngsp.org.

6. Detection Limit:

The claimed measuring range for the HbA1c values of the Hipro glycosylated hemoglobin (HbA1c) test kit is 4.3 to 14%.

7. Assay Cut-Off:

Not applicable.

8. Accuracy (Instrument):

Not applicable. See method comparison study below.

9. Carry-Over:

Not applicable.

B. Comparison Studies:

1. Method Comparison with Predicate Device:

Method comparison testing was performed using 120 human whole blood samples in K2 EDTA with values spanning the assay measurement range of HbA1c. The candidate device results were compared to those from testing performed at a secondary NGSP reference laboratory on the Bio-Rad D-100 HbA1c Test as the comparator method. The distribution of samples spanning the measuring interval is as follows:

% HbA1c	N	% of sample tested
≤ 5%	9	7.5
5.0 – 6.0%	11	9.2
6.0 – 6.5%	25	20.8
6.5 – 7.0%	36	30
7.0 – 8.0%	10	8.3
8.0 – 9.0%	10	8.3
> 9.0%	19	15.8
Total	120	100

Regression analyses were performed for the candidate device versus the comparator method. Summary of regression results are as follows:

Method	Slope	95% CI	y-intercept	95% CI	R
Linear	1.089	0.9952, 1.0226	-0.0727	-0.1775, 0.0321	0.9972
Deming	0.9884	0.9722, 1.0044	0.0926	-0.0188, 0.2040	0.9972
Passing-Bablok	1.000	1.000, 1.0256	0.0000	-0.1782, 0.0000	0.9830

Bias Estimation:

%HbA1c	Bias	%Bias
5.23%	-0.0262	-0.5%
6.34%	-0.0163	-0.26%
8.03%	-0.0012	-0.02%
12.53%	0.0388	0.31%

Total Error:
The bias estimation values determined in the method comparison study and the precision determined in the precision study were used to determine the total error at each of the HbA1c levels listed in the tables below. Total error was calculated by the following equation:

$$\%TE = |\%Bias| + (1.96 \times \%CV) \times (1 + \%Bias/100)$$

HbA1c Concentration, %	%Bias	%CV	%TE
5.23	-0.5%	2.0	4.4
6.34	-0.26%	2.7	5.5
8.03	-0.02%	2.4	4.7
12.53	0.31%	1.5	3.3

2. Matrix Comparison:

   Not applicable.

C Clinical Studies:

1. Clinical Sensitivity:

   Not applicable

2. Clinical Specificity:

   Not applicable

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

   Not applicable

D Clinical Cut-Off:

Not applicable.

E Expected Values/Reference Range:

The following HbA1c ranges recommended by the American Diabetes Association (ADA)[1-3] may be used as an aid in the diagnosis of diabetes mellitus:

HbA1c% (NGSP)	Suggested Diagnosis
Less than 5.7	Non-Diabetic
5.7 to 6.4	Pre-Diabetic
6.5 or higher	Diabetic

The expected HbA1c range for non-diabetic adults is 4.0-6.0 %[4]

1. Hemoglobin A1C, Emily Eyth; Roopa Naik. StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024 Jan. 2023 Mar 13.
2. American Diabetes Association. 6. Glycemic Targets: Standards of Medical Care in Diabetes-2019. Diabetes Care. 2019 Jan;42(Suppl 1):S61-S70.
3. American Diabetes Association. Diagnosis and Classification of Diabetes Mellitus. Diabetes Care 2010, 33 (Suppl. 1), S62-S69.
4. HbA1c: A Review of Analytical and Clinical Aspects, Cas Weykamp, Ph.D., Ann Lab Med 2013;33:393-400.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

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