510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:
k102908

B. Purpose for Submission:
Clearance for new device

C. Measurand:
Factor IX activity

D. Type of Test:
Quantitative

E. Applicant:
r² Diagnostics, Inc

F. Proprietary and Established Names:
NoFact IX Deficient Plasma

G. Regulatory Information:

1. Regulation section:
   21 CFR § 864.7290, Factor deficiency test
2. Classification:
   Class II
3. Product code:
   GJT, Plasma, Coagulation factor deficient
4. Panel:
   81 Hematology

H. Intended Use:

1. Intended use(s):
   NoFact IX Deficient Plasma is a human plasma immunodepleted of Factor IX and intended for the quantitative determination of Factor IX activity in citrated plasma from patients suspected of FIX deficiency. FIX activity is based on the activated partial thromboplastin time. For *in vitro* diagnostic use.
2. Indication(s) for use:
   Same as Intended use
3. Special conditions for use statement(s):
   For prescription use only
4. Special instrument requirements:
   Diagnostica Stago STA Family analyzers

I. Device Description:
The r² NoFact IX Deficient plasma kit contains 10 x 1 mL vials of lyophilized human plasma artificially immunodepleted of Factor IX to contain less than 1% factor IX activity with buffer and stabilizers.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   STAGO Factor IX-Deficient Plasma
2. Predicate 510(k) number(s):
   k933432
3. Comparison with predicate:

Similarities		
Item	Device: r² NoFact IX Deficient Plasma (k102908)	Predicate: STAGO Factor IX-Deficient Plasma (k933432)
Intended Use	NoFact IX Deficient Plasma is a human plasma immunodepleted of Factor IX and intended for the quantitative determination of Factor IX activity in citrated plasma.	Same
Measurement Principle	Clot-based assay for Factor IX activity using a modified activated partial thromboplastin time (APTT) test and Factor IX deficient plasma.	Same
Format	Lyophilized plasma	Same
Analyte tested	Factor IX activity	Same

Differences		
Item	Device:r² NoFACT IX Deficient Plasma (k102908)	Predicate: STAGO Factor IX-Deficient Plasma (k933432)
Reconstituted Stability	2-8ºC: 8hrs Room Temperature (18º-26ºC): 4 hrs	2-8ºC: 8hrs Room Temperature: not listed On-board STA Compact® and STA-R®: 4hrs
Final Product Form	Kit contains 10 x 1 mL lyophilized vials	Kit containing 6 x 1 mL lyophilized vials

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

CLSI EP5-A2, Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guidelines, Second Edition, 2004.

CLSI EP6-A, Evaluation of the Linearity of Quantitative Measurement Procedures: A statistical Approach; Approved Guidelines, 2003.

CLSI EP9-A2 Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline, Second Edition, 2002.

CLSI EP25-A, Evaluation of Stability of In Vitro Diagnostic Reagents; Approved Guideline, 2009.

CLSI H21-A5, Collection, Transport and Processing of Blood Samples for Testing Plasma-based Coagulation Assays and Molecular Hemostasis Assays; Approved Guideline-Fifth Edition, 2008.

L. Test Principle:

The assay determines the functional activity of Factor IX by measuring the degree of prolongation of activated partial thromboplastin time in the presence of a contact activator, thromboplastin, phospholipids and calcium ions. Factor IX activity is correlated with the prolongation of the clotting time of the Factor IX deficient plasma when added to diluted patient plasma. The APTT clotting time of this mixture is inversely proportional to factor IX concentration (% activity) in the patient plasma.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Studies for within-run, lot-to-lot and within-device precision were performed on low % activity factor IX pooled plasma for 10 days along with normal and high controls for 20 days. The controls and low % activity pooled plasma were tested with 3 lots of Stago PTT-A reagent and 3 lots of NoFACT IX deficient plasma on the Stago STA Compact analyzer. Testing was conducted twice a day with two replicates per run for each sample. The acceptance criteria for within-run and lot-to-lot %CV is <10% and for within device %CV is <15%. The overall results of the study met the acceptance criteria as follows:

Sample Type	Mean FVIII activity %	% CV Lot-to-Lot	NoFact VIII Lot#	% CV Between - day	% CV Within-run	% CV Within-Device
System N (Normal Control Plasma)	109%	1.1%	1	3.2	5.3	6.6
			2	4.4	4.4	7.0
			3	2.6	5.4	7.6
System P (Abnormal Control Plasma)	43%	2.9%	1	4.4	5.2	7.3
			2	4.0	4.1	7.2
			3	3.5	4.4	6.7

b. Linearity/assay reportable range:

Linearity studies were performed using the Stago PTT-A reagent on the STA Compact analyzer. The assays were calibrated using the Stago Unicalibrator. A series of 11 dilutions was conducted from reconstituted ISTH SSC approved plasma and diluted with the assay diluent and was tested at concentrations ranging from 0.8 to 188% activity. Duplicate measurements of each dilution were obtained with each of 3 lots of NoFACT IX. The Factor IX dilutions were run in duplicate for each NoFACT VIII Deficient Plasma lot and the average activities were plotted against their assigned values The acceptance criteria were as follows: slope of 0.8-1.2, intercept <6.0 and r >0.9. The calculated slope, intercept, and correlation coefficient are provided in the following table:

NoFact IX Deficient Plasma	Slope	Intercept	r-value
Lot 1	1.212	-0.57	0.9998
Lot 2	1.084	1.168	0.9959
Lot 3	1.066	0.815	0.9979

The results of the study were within acceptable limits.

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

Real-time Stability:

Three lots of NoFACT IX Deficient Plasma were tested with Stago System N and P control plasmas for factor IX recovery for real-time stability. The lots were tested on the Stago STA Compact using Stago PTT-A, Stago System N

and P control plasmas, and Stago Unicalibrator. The NoFact IX plasma was stored, reconstituted and tested from 0 months through 33 months at specific time intervals. Factor IX recovery of all three lots was acceptable based on the established reference range of each control. Results for the accelerated stability study, which projects a 2.4 year shelf life, support a 2-year shelf life claim of NoFact IX Deficient Plasma when stored at 2-8 °C. Ongoing real-time stability testing will be used to update the shelf life.

Reconstituted stability at Room Temperature and 2-8ºC:

Two lots of NoFact IX Deficient Plasma were reconstituted according to the package insert. The Stago PTT-A and NoFact IX deficient plasma assay was used to test the System N and P controls for factor IX recovery. The recovered FIX activity values at the time of reconstitution were considered the "zero hour" baseline. The reconstituted vials were stored at 0, 2 ,6, 8 and 24 hours at 2-8ºC and 0, 4, 6 and 8 hours at room temperature. At the time periods after reconstitution, vials from both sets of temperatures were run and the recovery at that time point was determined. These recoveries were compared to the baseline recovery by calculating the % shift in value: (baseline value - time point value) divided by (baseline value). The acceptance criterion is a shift of ≤10% compared to the zero hour baseline. The overall results meet the acceptance criteria with study results at room temperature of <8.1% and 2-8ºC at <7.5%. Stability data supports the reconstituted stability claim of 8 hours at 2-8ºC and 4 hours at room temperature.

d. Detection limit:

Not applicable

e. Analytical specificity:

Interference studies were conducted using pooled normal plasma (PNP) spiked with the interferents at predetermined maximum concentrations to create a stock preparation. The stock preparation was serially diluted 12 times with PNP in which each dilution was tested for factor IX activity on the STA Compact using Stago PTT-A and NoFACT IX. The % difference between the spiked dilutions to baseline (PNP) was accessed with an overall % difference <6%, which met the acceptance criteria difference of <10%.

Maximum Concentration results are as follows:

Interferents	Maximum Concentration
Hemolysis	500 mg/dL
Icterus	20 mg/dL
Lipemia	2000 mg/dL (triglycerides)
Low Molecular Weight Heparin	5 anti-Xa units/mL
Direct Thrombin Inhibitor	20 ug/mL
Unfractionated Heparin	2.0 Units/mL

f. Assay cut-off:

Not applicable

2. Comparison studies:

a. Method comparison with predicate device:

NoFact IX Deficient Plasma was compared to the predicate STA IX Deficient plasma using the Stago PTT-A with factor IX deficient plasma assay on the

STA Compact measuring the factor IX % activity range from 6% to 220%. A total of 233 (124 normal/109 abnormal) donors and patient samples, were analyzed over three laboratory sites. The clinical distribution of the samples is detailed in the tables below:

Table 1:

Gender	Sample size (n)	Age	Sample size (n)
Male	147	<21 yrs	22
Female	86	21-60 yrs	193
		>60 yrs	18

Table 2:

	Site 1	Site 2	Site 3
Total (n):	100	80	53
Normal donors:	45	44	44
Abnormal:	55	36	9
- Hemophilia	50	28	7
- Sepsis	2	2	1
- VWD	3	4	-
- Surgery	-	2	-
- Other	-	-	1

Citrated plasma samples were tested in parallel with the Stago PTT-A assay using the STA FIX and NoFact IX deficient plasma. The acceptance criteria for the study were: slope between 0.8-1.2, and correlation coefficient (r) >0.90. Data analysis for the overall study is as follows:

Table 3:

All Sites (n = 233)	
Slope (95% CI)	0.858 (0.824-0.892)
Intercept (95% CI)	5.729 (2.175-9.238)
Correlation coefficient (r)	0.956

b. Matrix comparison:

Fresh vs. Frozen (Thawed) -Testing was performed on the Stago Compact using Stago PTT-A assay and NoFact IX Deficient Plasma. A total of 18 (9 normal & 9 abnormal) samples were tested. The samples were split, tested fresh and then frozen at 20ºC and 80ºC for 1-3 days and later thawed and re-tested. A linear regression was conducted from the test results from the samples thawed from -20ºC and -80ºC. The acceptance criterion is r >0.90. The slope, intercept and correlation coefficient (r) at -20ºC were 1.065, -0.02 and 0.994 respectively. The slope, intercept, and correlation coefficient (r) for

the -80ºC results were 0.995, -0.02, and 0.997 respectively. The study results met the acceptance criterion.

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:
   To verify the normal range for factor IX activity, a total of 109 normal samples were tested for factor IX activity using Stago PTT-A assay and NoFact IX deficient plasma on the Stago Compact. The data was used to calculate a 2SD distribution for the normal range activity for factor IX. The results verified the cited literature normal range of 78%-184%.

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.