DEPARTMENT OF HEALTH & HUMAN SERVICES Public Health Service

Food and Drug Administration
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Silver Spring, MD 20993-0002

September 8, 2017

Instrumentation Laboratory Co.
Carol Marble
Regulatory Affairs Director
180 Hartwell Road
Bedford, Massachusetts 01730

Re: K170854
Trade/Device Name: HemosIL AcuStar HIT-IgG$_{(PF4-H)}$, HemosIL AcuStar HIT Controls
Regulation Number: 21 CFR 864.7695
Regulation Name: Platelet factor 4 radioimmunoassay
Regulatory Class: Class II
Product Code: LCO, GGN
Dated: March 21, 2017
Received: March 22, 2017

Dear Carol Marble:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration. Please note: CDRH does not evaluate information related to contract liability warranties. We remind you, however, that device labeling must be truthful and not misleading.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR

Part 807); labeling (21 CFR Part 801 and Part 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820); and if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801 and Part 809), please contact the Division of Industry and Consumer Education (DICE) at its toll-free number (800) 638-2041 or (301) 796-7100 or at its Internet address http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm for the CDRH's Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Industry and Consumer Education (DICE) at its toll-free number (800) 638-2041 or (301) 796-7100 or at its Internet address http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm.

Sincerely,

Leonthena R. Carrington -S

Lea Carrington
Director
Division of Immunology
and Hematology Devices
Office of In Vitro Diagnostics
and Radiological Health
Center for Devices and Radiological Health

Enclosure

DEPARTMENT OF HEALTH AND HUMAN SERVICES
Food and Drug Administration

Indications for Use

Form Approved: OMB No. 0910-0120
Expiration Date: 06/30/2020
See PRA Statement below.

510(k) Number *(if known)*
K170854

Device Name
HemosIL AcuStar HIT-IgG(PF4-H)
HemosIL AcuStar HIT Controls

Indications for Use *(Describe)*

HemosIL AcuStar HIT-IgG(PF4-H) is a qualitative, fully automated, chemiluminescent immunoassay (CIA) for the detection of IgG antibodies that react with Platelet Factor 4 (PF4) when complexed to heparin. The assay is for use in human 3.2% or 3.8% citrated plasma and serum on the ACL AcuStar instrument in a laboratory setting.

The result provided by the assay should be interpreted as either positive or negative based on the assay cut-off (1.00 U/mL). The positive or negative result aids in determining the risk for heparin induced thrombocytopenia (HIT) when used in conjunction with other laboratory and clinical findings.

Anti-PF4/Heparin antibodies are commonly found in patients with HIT. For use in adult population suspected of HIT. Not for use in isolation to exclude HIT.

HemoslL AcuStar HIT Controls are for the quality control of the HemosIL AcuStar HIT-IgG(PF4-H) assay as performed on the ACL AcuStar.

For prescription use.

Type of Use *(Select one or both, as applicable)*

☒ Prescription Use (Part 21 CFR 801 Subpart D) ☐ Over-The-Counter Use (21 CFR 801 Subpart C)

CONTINUE ON A SEPARATE PAGE IF NEEDED.

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Instrumentation Laboratory
A Werfen Company

510(k) Summary

This 510(k) Summary of Safety and Effectiveness is being submitted in accordance with the requirements of the Safe Medical Device Act of 1990 and 21 CFR 807.92.

Submitter's Information	Instrumentation Laboratory (IL) Co. 180 Hartwell Road Bedford, MA 01730, USA	
Contact Person	Carol Marble, Regulatory Affairs Director Phone: 781-861-4467 Fax: 781-861-4207 Email: cmarble@ilww.com	
Preparation Date	August 26, 2017	
Device Trade Names	Reagent Cartridge Kit (with Calibrator)	HemosIL AcuStar HIT-IgG$_{(PF4\text{-}H)}$
	Controls	HemosIL AcuStar HIT Controls
Regulatory Information – Reagent Cartridge Kit	Regulation Number	21 CFR 864.7695
	Regulation Description	Platelet Factor 4 Radioimmunoassay
	Classification	Class II
	Product Code	LCO
	Classification Panel	Hematology (81)
Regulatory Information – Controls	Regulation Number	21 CFR 864.5425
	Regulation Description	Plasma, Coagulation Control
	Classification	Class II
	Product Code	GGN
	Classification Panel	Hematology (81)
Predicate Device	K071255 (Zymutest HIA IgG from HYPHEN Biomed)	

Indications for Use / Intended Use	HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ is a qualitative, fully automated, chemiluminescent immunoassay (CIA) for the detection of IgG antibodies that react with Platelet Factor 4 (PF4) when complexed to heparin. The assay is for use in human 3.2% or 3.8% citrated plasma and serum on the ACL AcuStar instrument in a laboratory setting. The result provided by the assay should be interpreted as either positive or negative based on the assay cut-off (1.00 U/mL). The positive or negative result aids in determining the risk for heparin induced thrombocytopenia (HIT) when used in conjunction with other laboratory and clinical findings. Anti-PF4/Heparin antibodies are commonly found in patients with HIT. For use in adult population suspected of HIT. Not for use in isolation to exclude HIT. HemosIL AcuStar HIT Controls are for the quality control of the HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay as performed on the ACL AcuStar. For prescription use.
Device Description **Reagent Cartridges with Calibrators**	The HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay is a chemiluminescent two-step immunoassay consisting of magnetic particles coated with PF4 complexed to polyvinyl sulfonate (PVS) which capture, if present, the PF4/Heparin antibodies from the sample. After incubation, magnetic separation, and a wash step, a tracer consisting of an isoluminol-labeled anti-human IgG antibody is added and may bind with the captured PF4/Heparin IgG on the particles. After a second incubation, magnetic separation, and a wash step, reagents that trigger the luminescent reaction are added, and the emitted light is measured as relative light units (RLUs) by the ACL AcuStar optical system. The RLUs are directly proportional to the PF4/Heparin IgG concentration in the sample. The **HemosIL AcuStar HIT-IgG$_{(PF4-H)}$** kit consists of: R **HIT-IgG$_{(PF4-H)}$ Cartridge for 25 determinations**: Cartridge containing 1 vial of magnetic particle suspension coated with PF4/PVS complex, 1 vial of assay buffer, 1 vial of tracer consisting of an mAb anti-human IgG antibody labeled with isoluminol, and 1 vial of sample diluent used for the regular predilution of the sample. The reagents are in a phosphate or Tris buffer containing bovine serum albumin, bovine fetal serum, PF4/PVS complex, mouse monoclonal IgG, stabilizers, and preservative. C1 **HIT-IgG$_{(PF4-H)}$ Calibrator 1**: Barcoded tube of a solution with humanized mAb anti-PF4-Heparin in Tris buffer containing bovine serum albumin, stabilizers and preservative. C2 **HIT-IgG$_{(PF4-H)}$ Calibrator 2**: Barcoded tube of a solution with humanized mAb anti-PF4-Heparin in Tris buffer containing bovine serum albumin, stabilizers, and preservative. The calibrators are lot specific and they cannot be used with other lots of reagents.
Controls	The Low and High HIT Controls are prepared by means of a dedicated process and contain different concentrations of humanized monoclonal anti-PF4-Heparin. **Low HIT Control:** Control intended for the assessment of precision and accuracy of the HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay below the cut-off. **High HIT Control:** Control intended for the assessment of precision and accuracy of the HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay above the cut-off. Use of both controls is recommended for a complete quality control program.

Comparison to Predicate		
Item	**Predicate**	**New Device**
Trade Names	Zymutest HIA IgG (Kit Includes 2 Control Levels)	HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ HemosIL AcuStar HIT Controls
Manufacturer	HYPHEN Biomed	Instrumentation Laboratory Co.
Similarities		
Measurand	Anti-PF4/Heparin IgG Antibodies	Same
Assay Type	Qualitative	Same
Product Code	LCO	Same
Regulation Section	864.7695	Same
Classification	Class II	Same
Intended Use	The ZYMUTEST HIA, IgG ELISA kit, is a qualitative assay intended for the detection of heparin-dependent antibodies of the IgG isotype, in human plasma, by clinical laboratories. It is intended for in vitro diagnostic use.	HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ is a qualitative, fully automated, chemiluminescent immunoassay (CIA) for the detection of IgG antibodies that react with Platelet Factor 4 (PF4) when complexed to heparin. The assay is for use in human 3.2% or 3.8% citrated plasma and serum on the ACL AcuStar instrument in a laboratory setting. The result provided by the assay should be interpreted as either positive or negative based on the assay cut-off (1.00 U/mL). The positive or negative result aids in determining the risk for heparin induced thrombocytopenia (HIT) when used in conjunction with other laboratory and clinical findings. Anti-PF4/Heparin antibodies are commonly found in patients with HIT. For use in adult population suspected of HIT. Not for use in isolation to exclude HIT. HemosIL AcuStar HIT Controls are for the quality control of the HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay as performed on the ACL AcuStar. For prescription use.

Differences		
Sample Type	Citrated Human Plasma	Citrated Human Plasma or Serum
Methodology	Enzyme-linked immunosorbent assay (ELISA)	Chemiluminescent immunoassay (CIA)
Instrumentation	Manual	Automated ACL AcuStar instrument
Reagents	Microtiter plate coated with unfractionated heparin	Cartridge containing magnetic particle suspension coated with PF4 complexed to polyvinyl sulfonate (PVS)
Antibodies	Goat antibodies specific for human IgG	Mouse monoclonal anti-human IgG antibody
Conjugate	Peroxidase conjugated anti-human IgG	Isoluminol conjugated anti-human IgG
Cut-off	When the assay is run at 20±1°C, the results are as follows: • Positive: A450 > 0.50 • Weakly Positive: A450 > 0.30 to < 0.50 • Negative: A450 ≤ 0.30	Fixed clinical cut-off: • Positive: ≥ 1.00 U/mL • Negative: < 1.00 U/mL
Controls	Controls included in test kit: • Negative level • Positive level Lyophilized Format	Controls sold separately: • Low Level below the cut-off • High Level above the cut-off Liquid Format
Calibration	Not Applicable	Lot specific Master Curve + two Calibrators (included in kit) Liquid Format

Performance Summary

Multi-Reagent Cartridge Lot Precision

An internal precision study was performed using three (3) different lots of HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ reagent cartridges run on an ACL AcuStar. The study used five (5) plasma samples to span the assay range; two native (unadulterated) patient samples and three prepared patient sample pools.

Each material was tested with each reagent lot in duplicate, twice a day for 22 days, for a total of 88 replicates per level per lot as summarized below:

Reagent Lot No. 1			
Material	**Mean (U/mL)**	**Within Run (Repeatability) % CV**	**Total (Within Device) % CV**
Plasma Sample 1 (Pool)	0.62	8.3%	8.3%
Plasma Sample 2 (Pool)	2.23	6.5%	6.8%
Plasma Sample 3 (Native)	21.04	4.8%	5.6%
Plasma Sample 4 (Native)	46.15	4.7%	4.8%
Plasma Sample 5 (Pool)	90.35	4.4%	4.4%
Reagent Lot No. 2			
Material	**Mean (U/mL)**	**Within Run (Repeatability) % CV**	**Total (Within Device) % CV**
Plasma Sample 1 (Pool)	0.52	8.4%	9.2%
Plasma Sample 2 (Pool)	1.98	7.5%	9.3%
Plasma Sample 3 (Native)	26.13	6.1%	6.1%
Plasma Sample 4 (Native)	54.88	6.9%	7.5%
Plasma Sample 5 (Pool)	111.74	4.1%	7.3%
Reagent Lot No. 3			
Material	**Mean (U/mL)**	**Within Run (Repeatability) % CV**	**Total (Within Device) % CV**
Plasma Sample 1 (Pool)	0.49	13.0%	13.8%
Plasma Sample 2 (Pool)	1.69	7.1%	7.4%
Plasma Sample 3 (Native)	18.40	7.4%	8.1%
Plasma Sample 4 (Native)	36.85	4.8%	5.6%
Plasma Sample 5 (Pool)	86.79	6.8%	7.4%

Aggregated data (Reagent Lots 1, 2 and 3)		
Material	**Mean (U/mL)**	**Lot-to-Lot Variability %CV**
Plasma Sample 1 (Pool)	0.54	11.8%
Plasma Sample 2 (Pool)	1.97	13.8%
Plasma Sample 3 (Native)	21.85	18.0%
Plasma Sample 4 (Native)	45.96	19.6%
Plasma Sample 5 (Pool)	96.30	14.0%

Cut-off Precision

An additional internal precision study was performed with three (3) different lots of HemosIL AcuStar HIT-IgG$_{(PF4\text{-}H)}$ reagent cartridges, using three (3) plasma samples at levels near the claimed assay cut-off (1: negative sample pool; 2: positive sample pool; 3: unadulterated positive sample).

Each sample was analyzed with each of the three (3) reagent lots on two (2) different ACL AcuStar instruments by two (2) different operators over five (5) days for two (2) runs per day in three (3) replicates per run (N=360 per sample level) as summarized below:

Sample	N	Mean	Within-Run (SD, %CV)	Between-Run (SD, %CV)	Between-Day (SD, %CV)	Between-Lot (SD, %CV)	Between-Instrument (SD, %CV)	Between-Operator (SD, %CV)	Total (SD, %CV)
1	360	0.79	0.04; 4.7%	0.02; 2.1%	0.02; 2.0%	0.07; 8.3%	0.00; 0.0%	0.00; 0.0%	0.08; 10.0%
2	360	1.43	0.06; 4.3%	0.04; 3.0%	0.00; 0.0%	0.07; 4.8%	0.00; 0.0%	0.00; 0.0%	0.10; 7.1%
3	360	3.61	0.13; 3.6%	0.14; 3.7%	0.05; 1.5%	0.12; 3.4%	0.00; 0.0%	0.00; 0.0%	0.23; 6.4%

Multi-Control Lot Precision

An internal precision study was performed using three (3) different lots of HemosIL AcuStar HIT Controls (low and high) run on an ACL AcuStar, with a single lot of HemosIL AcuStar HIT-$IgG_{(PF4\text{-}H)}$.

Each level of control material from each lot was tested in duplicate, twice a day for 20 days, for a total of 80 replicates per level per lot as summarized below:

Material	Mean (U/mL)	Within Run (Repeatability) % CV	Total (Within Device) % CV
Low HIT Control Lot No. 1	0.58	4.6%	7.0%
High HIT Control Lot No. 1	3.16	3.5%	6.1%
Low HIT Control Lot No. 2	0.49	3.2%	7.0%
High HIT Control Lot No. 2	2.77	3.4%	5.0%
Low HIT Control Lot No. 3	0.60	4.1%	6.3%
High HIT Control Lot No. 3	2.96	2.8%	5.5%

Multi-Calibrator Lot Precision

An internal precision study was performed using three (3) different lots of HemosIL AcuStar HIT-$IgG_{(PF4\text{-}H)}$ Calibrator (kit component from 3 different assay lots) run on an ACL AcuStar, with a single lot of HemosIL AcuStar HIT-$IgG_{(PF4\text{-}H)}$ (with a different kit Calibrator lot).

Each calibrator lot was tested in duplicate, twice a day for 20 days, for a total of 80 replicates per lot as summarized below:

Material	Mean (U/mL)	Within Run (Repeatability) % CV	Total (Within Device) % CV
Kit Calibrator 1 Lot No. 1	0.92	3.9%	6.3%
Kit Calibrator 2 Lot No. 1	15.37	3.2%	3.9%
Kit Calibrator 1 Lot No. 2	0.94	2.7%	5.4%
Kit Calibrator 2 Lot No. 2	14.85	2.2%	3.1%
Kit Calibrator 1 Lot No. 3	0.90	2.4%	4.0%
Kit Calibrator 2 Lot No. 3	14.90	3.3%	5.0%

Multi-Reagent Cartridge and Control Lot Reproducibility

Reproducibility studies were conducted at three (3) external clinical sites by three (3) different operators (one operator per site), on three (3) different ACL AcuStar instruments (one instrument per site), using three (3) different lots of HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ reagent cartridges and HemosIL AcuStar HIT Controls (low and high). The same three (3) patient citrated plasma sample pools (2 positive and 1 negative) were also tested across the three (3) sites.

Each material was tested in triplicate, twice a day for 5 days, for a total of 30 replicates per level.

The pooled data for each reagent lot are summarized below:

Pooled 3 Site Data: Reagent Lot No. 1 of HemosIL AcuStar HIT-IgG$_{(PF4-H)}$												
Level	**Mean (U/mL)**	**N**	**Repeatability (within-run)**		**Between-Run**		**Between-Day**		**Between Site**		**Reproducibility (Total)**	
			SD	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**
Low HIT Control	0.64	90	0.02	2.5	0.03	4.7	0.01	1.7	0.06	9.2	0.07	10.7
High HIT Control	3.05	90	0.10	3.3	0.10	3.2	0.05	3.2	0.27	8.8	0.32	10.5
Plasma Sample 1	0.06	90	All Replicates < 1.00 U/mL									
Plasma Sample 2	3.52	90	0.17	4.8	0.09	2.7	0.07	1.8	0.16	4.6	0.26	7.4
Plasma Sample 3	21.33	90	1.53	7.2	0.00	0.0	0.37	1.7	0.77	3.6	1.75	8.2

Pooled 3 Site Data: Reagent Lot No. 2 of HemosIL AcuStar HIT-IgG$_{(PF4-H)}$												
Level	**Mean (U/mL)**	**N**	**Repeatability (within-run)**		**Between-Run**		**Between-Day**		**Between Site**		**Reproducibility (Total)**	
			SD	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**
Low HIT Control	0.56	90	0.02	3.6	0.02	2.9	0.02	3.9	0.03	5.7	0.05	8.2
High HIT Control	2.66	90	0.11	4.3	0.09	3.2	0.06	2.2	0.14	5.3	0.21	7.8
Plasma Sample 1	0.04	90	All Replicates < 1.00 U/mL									
Plasma Sample 2	3.23	90	0.15	4.6	0.14	4.4	0.10	3.1	0.24	7.3	0.33	10.2
Plasma Sample 3	20.79	90	1.41	6.8	0.42	2.0	0.50	2.4	0.88	4.2	1.79	8.6

Pooled 3 Site Data: Reagent Lot No. 3 of HemosIL AcuStar HIT-IgG$_{(PF4-H)}$												
Level	**Mean (U/mL)**	**N**	**Repeatability (within-run)**		**Between-Run**		**Between-Day**		**Between Site**		**Reproducibility (Total)**	
			SD	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**	**SD**	**%CV**
Low HIT Control	0.47	90	0.02	3.6	0.02	4.6	0.02	3.2	0.01	2.7	0.03	7.2
High HIT Control	2.62	90	0.10	3.9	0.11	4.4	0.12	4.5	0.06	2.3	0.20	7.7
Plasma Sample 1	0.06	90	All Replicates < 1.00 U/mL									
Plasma Sample 2	4.47	90	0.19	4.3	0.28	6.3	0.21	4.7	0.08	1.8	0.41	9.2
Plasma Sample 3	29.64	90	1.84	6.2	1.54	5.2	1.80	6.1	0.91	3.1	3.14	10.6

Result Interpretation

HIT-IgG$_{(PF4-H)}$ results are reported in U/mL (HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ Arbitrary Units) on the ACL AcuStar instrument as follows:

System	Results	Interpretation
ACL AcuStar	$\geq$ 1.00 U/mL	HIT IgG Antibody positive
ACL AcuStar	< 1.00 U/mL	HIT IgG Antibody negative

IMPORTANT: The laboratory should report only the interpreted result (HIT Antibody positive or negative) to the clinician as the final reported result.

The positive or negative result should be used with other information, including the clinical context, in forming a diagnosis such as the 4T score and the 2013 American Society of Hematology guidelines.

Although a positive result obtained using this assay may indicate the presence of heparin-associated antibodies, a positive result **DOES NOT CONFIRM** the diagnosis of HIT. Some patients may have naturally occurring antibodies to PF4.

Interferences / Limitations

- **Interferences**

  Testing confirmed no interference for HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ on the ACL AcuStar up to the following concentrations:

Hemoglobin	Bilirubin	Triglycerides	Heparin LMW and UF	HAMA
500 mg/dL	18 mg/dL	1250 mg/dL	1 IU/mL	1 µg/mL

  Note: The presence of Rheumatoid Factor may produce an over estimation of the test result with the HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay.

- **Antiphospholipid Syndrome (APS)**

  Twenty six (26) citrated plasma samples from patients diagnosed with Antiphospholipid Syndrome (APS) were tested with HemosIL AcuStar HIT-IgG$_{(PF4-H)}$. All 26 samples reported as negative with HemosIL AcuStar HIT-IgG$_{(PF4-H)}$, demonstrating that the assay is not affected by APS antibodies

Cut-Off Determination

Eighty-seven (87) citrated plasma samples were obtained from hospitalized patients who had been exposed to heparin, and who displayed clinical signs consistent with Heparin Induced Thrombocytopenia (HIT). These patients were tested by the hospital with the Serotonin Release Assay (SRA) and the sample results (45 SRA positive and 42 SRA negative) were used to perform a Receiver Operating Characteristics (ROC) analysis. The cut-off was established as 1.00 U/mL (94.3% Agreement; 95% CI = 87.2% - 97.5%).

Reference Interval – Heparin Exposed, Non-HIT Suspected Patients

A population of 91 citrated plasma samples from heparin exposed, but non-HIT suspected patients was tested. The non-parametric 95% reference interval was calculated and the upper limit determined to be 0.75 U/mL (90% CI: 0.46 to 2.86 U/mL).

Reference Interval – Healthy Donors

A population of 154 citrated plasma samples from apparently healthy individuals was tested. The non-parametric 95% reference interval was calculated and the upper limit determined to be 0.37 U/mL (90% CI: 0.17 to 1.25 U/mL).

Method Comparison Definitions and Formulas

See below 2x2 contingency table with definitions and formulas.

		Reference	
		+	-
Test	+	TP	FP
	-	FN	TN

TP	*True Positives*
TN	*True Negatives*
FP	*False Positives*
FN	*False Negatives*

PPA Positive Percent Agreement
NPA Negative Percent Agreement

$$PPA = \frac{TP}{TP + FN} \qquad NPA = \frac{TN}{TN + FP}$$

NPV Negative Predictive Value
PPV Positive Predictive Value

$$PPV = \frac{TP}{TP + FP} \qquad NPV = \frac{TN}{TN + FN}$$

TPA Total Percent Agreement

$$TPA = \frac{TP + TN}{TP + TN + FP + FN}$$

Multicenter Method Comparison

A multicenter method comparison study was performed at three (3) hospitals, comparing the performance of HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ with the predicate device, Zymutest HIA IgG (K071255) (n=802) and with the Serotonin Release Assay (SRA) (n=790).

The samples were from patients exposed to heparin that showed HIT related symptoms.

Samples were analyzed in singlicate, with no spiked samples used in this study.

- **HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ vs. Predicate Device (Zymutest HIA IgG)**

  The pooled results below are based on a cut-off of 1.00 U/mL for the HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay and 0.5 OD cut-off value for Zymutest HIA IgG.

		Zymutest HIA IgG Results		
		+	-	Total
HemosIL AcuStar HIT IgG$_{(PF4-H)}$ Results	+	26	9	35
	-	48	719	767
	Total	74	728	802

HemosIL AcuStar HIT IgG$_{(PF4-H)}$ vs. Zymutest HIA IgG	**Proportion**	**Wilson 95% CI**	
PPA (Positive Percent Agreement)	35% (26/74)	25%	47%
NPA (Negative Percent Agreement)	99% (719/728)	98%	99%
Total Percent Agreement	93% (745/802)	91%	94%

HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ Assay vs. SRA and Predicate vs. SRA

Patient samples in the multicenter study without a valid SRA result or with an indeterminate result (total N=12) were removed from calculations, bringing the final total to N = 790. The pooled results summarized below compare both HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ (cut-off of 1.00 U/mL) to the SRA test results and the Zymutest HIA IgG assay (cut-off of 0.5 OD) to the SRA test results.

- **HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ Assay vs. SRA:**

		SRA Results		
		+	-	Total
HemosIL AcuStar HIT IgG$_{(PF4-H)}$ Results	+	26	8	34
	-	15	741	756
	Total	41	749	790

HemosIL AcuStar HIT IgG$_{(PF4-H)}$ vs. SRA	**Proportion**	**Wilson 95% CI**	
PPV (Positive Predictive Value)	76% (26/34)	60%	88%
NPV (Negative Predictive Value)	98% (741/756)	97%	99%
Total Percent Agreement	97% (767/790)	96%	98%

- **Zymutest HIA IgG (Predicate) vs. SRA:**

		SRA Results		
		+	-	Total
Zymutest HIA IgG Results	+	26	47	73
	-	15	702	717
	Total	41	749	790

Zymutest HIA IgG vs. SRA	**Proportion**	**Wilson 95% CI**	
PPV (Positive Predictive Value)	36% (26/73)	26%	47%
NPV (Negative Predictive Value)	98% (702/717)	97%	99%
Total Percent Agreement	92% (728/790)	90%	94%

- **Conclusion for SRA testing:** HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ showed equivalent NPV and superior PPV performance to the predicate device when both assays were compared to the SRA method with the intended use adult population.

Conclusion:

The analytical and clinical study results demonstrate that the HemosIL AcuStar HIT-IgG$_{(PF4-H)}$ assay and HemosIL AcuStar HIT Controls are substantially equivalent to the predicate device, Zymutest HIA IgG (FDA cleared under K071255), and that the assay is safe and effective for its labeled intended use when compared to the SRA reference method.