

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY**

A. 510(k) Number:

k140686

B. Purpose for Submission:

New Device

C. Measurand:

Immunoglobulin IgM Kappa (combined μ heavy and κ light chain) and
Immunoglobulin IgM Lambda (combined μ heavy and λ light chain)

D. Type of Test:

Quantitative Immunoturbidmetric

E. Applicant:

The Binding Site Group, Ltd.

F. Proprietary and Established Names:

Hevylite™ Human IgM Kappa Kit for use on SPA_{PLUS}®
Hevylite™ Human IgM Lambda Kit for use on SPA_{PLUS}®

G. Regulatory Information:

1. Regulation section:
21 CFR § 866.5510, Immunoglobulins A, G, M, D, and E Immunological Test System
2. Classification:
Class II
3. Product code:
PDE – Immunoglobulin M kappa heavy and light chain combined
PDF – Immunoglobulin M lambda heavy and light chain combined
4. Panel:
Immunology 82

H. Intended Use:

1. Intended use(s):
Hevylite™ Human IgM Kappa Kit for use on SPA_{PLUS}®:

Hevylite Human IgM Kappa Kit for use on SPA_{PLUS} is intended for the in vitro quantification of IgM Kappa (combined μ heavy and κ light chain) concentration in human serum on the SPA_{PLUS}. The test result is to be used with previously diagnosed Waldenstrom's macroglobulinemia in conjunction with other clinical and laboratory findings.

This assay has not been established for the diagnosis, monitoring and prognosis of Waldenstrom's macroglobulinemia.

Hevylite™ Human IgM Lambda Kit for use on SPA_{PLUS}®:

Hevylite Human IgM Lambda Kit for use on SPA_{PLUS} is intended for the in vitro quantification of IgM Lambda (combined μ heavy and λ light chain) concentration in human serum on the SPA_{PLUS}. The test result is to be used with previously diagnosed Waldenstrom's macroglobulinemia in conjunction with other clinical and laboratory findings.

This assay has not been established for the diagnosis, monitoring and prognosis of Waldenstrom's macroglobulinemia.

2. Indication(s) for use:

Same as intended use.

3. Special conditions for use statement(s):

For prescription use only.

Warning: The result of Hevylite Human IgM Kappa in a given specimen determined with assays with different manufacturers can vary due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the Hevylite Human IgM Kappa assay used. Values obtained with different assay methods cannot be used interchangeably.

Warning: The result of Hevylite Human IgM Lambda in a given specimen determined with assays with different manufacturers can vary due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the Hevylite Human IgM Lambda assay used. Values obtained with different assay methods cannot be used interchangeably.

4. Special instrument requirements:

Binding Site SPA_{PLUS}®

I. Device Description:

The Hevylite™ Human IgM Kappa Kit and IgM Lambda Kit contain polystyrene latex coated with polyclonal monospecific sheep anti-human IgM antibody against combined μ heavy and κ light chain or combined μ heavy and λ light chain, on calibrator set (levels 1-6), two controls (low and high) and supplementary reagent buffer in liquid form. The reagents contain 0.099% sodium azide as preservative.

J. Substantial Equivalence Information:

1. Predicate device name(s) and 510(k) number(s):

Hevylite™ Human IgM Kappa Kit for use on Siemens BN™ II Systems, k113823
 Hevylite™ Human IgM Lambda Kit for use on Siemens BN™ II Systems, k113823

2. Comparison with predicate:

Similarities		
Item	Device Hevylite IgM Kappa and IgM Lambda kits use on SPA _{PLUS}	Predicate Hevylite IgM Kappa and IgM Lambda kits use on Siemens BN™ II Systems
Intended Use	Quantification of IgMκ (combined μ heavy and λ light chain) or IgMλ (combined μ heavy and λ light chain) concentration in human serum	Same
Indications for Use	Used with previously diagnosed Waldenstrom’s macroglobulinemia and in conjunction with other clinical and laboratory findings.	Same
Type of Test	Quantitative	Same
Sample Matrix	Serum	Same
Controls	One low and one high control (serum, ready to use)	Same
Calibrator	Binding Site Hevylite Calibrator™	Same
Antisera specificity	polyclonal monospecific sheep antibody (anti-human IgM combined μ heavy and κ light chain or combined μ heavy and λ light chain antiserum) coated onto polystyrene latex	Same
Open Vial stability	1 month	Same

Differences		
Item	Device Hevylite IgM Kappa and IgM Lambda kits use on SPA _{PLUS}	Predicate Hevylite IgM Kappa and IgM Lambda kits use on Siemens BN™ II Systems
Detection Method	Turbidimetric	Nephelometric
Instrument	Binding Site SPA _{PLUS}	BN™II
Measuring range	IgMκ: 0.2 – 5.0 g/L IgMλ: 0.18 – 4.5 g/L (at standard 1/10 dilution)	IgMκ: 0.2 – 6.4 g/L IgMλ: 0.175 – 5.60 g/L (at standard 1/100 dilution)

Differences		
Item	Device Hevylite IgM Kappa and IgM Lambda kits use on SPA _{PLUS}	Predicate Hevylite IgM Kappa and IgM Lambda kits use on Siemens BN TM II Systems
	Extended Range for IgMκ: 1/1 dilution: 0.02 – 0.50 g/L 1/10 dilution: 0.2 – 5.0 g/L 1/90 dilution: 1.8 – 45 g/L 1/250 dilution: 5 – 125 g/L Extended Range for IgMλ: 1/1 dilution: 0.018 – 0.450 g/L 1/10 dilution: 0.18 – 4.5 g/L 1/90 dilution: 1.620 – 40.5 g/L 1/250 dilution: 4.5 – 112.5 g/L	Extended Range for IgMκ: 1/5 dilution: 0.01 – 0.32 g/L 1/20 dilution: 0.04 – 1.28 g/L 1/100 dilution: 0.2 – 6.4 g/L 1/400 dilution: 0.8 – 25.6 g/L 1/2000 dilution: 4 – 128 g/L Extended Range for IgMλ: 1/5 dilution: 0.009 – 0.280 g/L 1/20 dilution: 0.035 – 1.12 g/L 1/100 dilution: 0.175 – 5.60 g/L 1/400 dilution: 0.7 – 22.4 g/L 1/2000 dilution: 3.5 – 112 g/L
Reference Range	IgMκ: 0.19 – 1.63 g/L IgMλ: 0.12 – 1.01 g/L IgM κ/λ ratio: 1.18 – 2.74 g/L	IgMκ: 0.29 – 1.82 g/L IgMλ: 0.17 – 0.94 g/L IgM κ/λ ratio: 0.96 – 2.3 g/L

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

CLSI EP-5A: Evaluation of Precision Performance of Clinical Chemistry.

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

CLSI EP-17A: Determination of Limits of Detection and Limits of Quantitation.

C28-A3: Defining, Establishing, and Verifying Reference Intervals in the Clinical Laboratory

EP07-A2: Interference Testing in Clinical Chemistry; Approved Guideline - Second Edition

L. Test Principle:

Evaluating the concentration of a soluble antigen (e.g. IgM Kappa and IgM Lambda) by turbidimetry involves the addition of the test sample (with either IgM Kappa (IgMκ) or IgM Lambda (IgMλ)) to a solution containing the appropriate antibody (anti-IgMκ or anti-IgMλ) in a reaction vessel or cuvette. A beam of light is passed through the cuvette and as the antigen-antibody reaction proceeds, the light passing through the cuvette is scattered increasingly as insoluble immune complexes are formed. Light scatter is monitored by measuring the decrease in intensity of the incident beam of light. The antibody in the cuvette is in excess so the amount of immune complex formed is proportional to the antigen concentration. A series of calibrators of known antigen concentration are assayed initially to produce a calibration curve of measured light scatter versus antigen concentration. Samples

of unknown antigen concentration can then be assayed and the results read from the calibration curve.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

This precision study was conducted in accordance with CLSI EP-05-A2 Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline - Second Edition. The study was carried out by testing three serum sample pools (high, medium and low) with different concentrations of IgMκ or IgMλ that span the measuring range of the assays at the recommended 1/10 dilution (0.2 – 5.0 g/L and 0.18 – 4.50 g/L respectively). The three samples were tested on three SPA_{PLUS} instruments and using three lots of assays over 21 days, with two runs per day in duplicate, for a total of 84 replicate measurements. The sponsor’s pre-defined acceptance criteria are total precision must be ≤ 10% for all levels, within-run precision ≤ 6%, and between-run and between-day ≤ 8% CV. Results are summarized below:

Hevylite™ Human IgM Kappa Kit for use on SPA_{PLUS}

IgMκ Sample	N	Mean g/L	Within-Run		Between-Run		Between-Day		Total Precision	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
High	84	4.13	0.08	1.8	0.08	1.8	0.21	4.6	0.24	5.3
Medium	84	1.80	0.03	1.5	0.03	1.3	0.66	3.5	0.08	4.1
Low	84	0.34	0.01	2.4	0.01	3.3	0.02	4.9	0.02	6.4

IgMκ Sample	N	Mean g/L	Between-Instrument		Between-lot	
			SD	%CV	SD	%CV
High	84	4.13	0.15	3.6	0.22	5.4
Medium	84	1.80	0.06	3.1	0.06	3.3
Low	84	0.34	0.01	3.8	0.00	0.3

Hevylite™ Human IgM Lambda Kit for use on SPA_{PLUS}:

IgMλ Sample	N	Mean g/L	Within-Run		Between-Run		Between-Day		Total Precision	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
High	84	4.11	0.07	1.7	0.06	1.5	0.18	4.4	0.20	5.0
Medium	84	0.96	0.02	1.9	0.01	0.6	0.03	3.2	0.03	3.8
Low	84	0.29	0.01	2.0	0.01	2.1	0.02	5.4	0.02	6.1

IgMλ Sample	N	Mean g/L	Between-Instrument		Between-lot	
			SD	%CV	SD	%CV
High	84	4.11	0.16	3.9	0.11	2.7
Medium	84	0.96	0.03	3.4	0.02	2.3
Low	84	0.29	0.02	5.0	0.01	3.3

In order to provide greater coverage of the assay measuring ranges (IgM Kappa: 0.2 – 5.0 g/L, IgM Lambda: 0.18 – 4.50 g/L), a second precision study was also performed according to CLSI EP05-A2. Five samples with concentrations covering the measuring range of IgM κ or IgM λ were tested on three SPA_{PLUS} instruments and using one lot of reagent over 21 days, with two runs per day in duplicate, for a total of 84 replicate measurements. The sponsor’s pre-defined acceptance criteria are total precision must be $\leq 10\%$ for all levels, within-run precision $\leq 6\%$, and between-run and between day $\leq 8\%$ CV. Results are summarized below:

IgM Kappa Precision Study:

IgM κ Sample	N	Mean g/L	Within-Run		Between-Run		Between-Day		Between-Instrument		Total Precision	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	84	0.34	0.01	1.8	0.01	2.7	0.01	3.2	0.01	2.7	0.02	4.6
2	84	1.13	0.16	1.4	0.03	2.3	0.02	1.9	0.02	1.9	0.03	3.3
3	84	1.89	0.28	1.8	0.04	2.3	0.07	4.6	0.07	3.6	0.08	5.4
4	84	4.50	0.04	0.9	0.07	1.5	0.09	2.1	0.07	1.6	0.12	2.8
5	84	5.20	0.05	1.0	0.11	2.1	0.14	2.5	0.07	1.3	0.19	3.5

IgM Lambda Precision Study:

IgM λ Sample	N	Mean g/L	Within-Run		Between-Run		Between-Day		Between-Instrument		Total Precision	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	84	0.26	0.01	2.0	0.01	3.3	0.01	4.1	0.01	4.7	0.01	5.7
2	84	0.71	0.01	1.3	0.02	3.0	0.00	0.0	0.01	0.8	0.02	3.3
3	84	1.22	0.01	1.0	0.07	5.8	0.00	0.0	0.02	2.0	0.07	5.9
4	84	3.86	0.05	1.3	0.16	4.2	0.10	2.6	0.14	3.5	0.20	5.1
5	84	5.24	0.06	1.1	0.34	6.5	0.19	3.6	0.33	6.3	0.39	7.5

b. Linearity/assay reportable range:

The linearity of the assays was assessed using one lot of reagent on one analyzer.

A high and a low pool were prepared for both the Hevylite Human IgM Kappa kit and the Hevylite IgM Lambda kit for use on SPA_{PLUS} in order to evaluate linearity across the measuring range. The high pools for IgM κ and IgM λ were prepared from a serum sample with a naturally high concentration of IgM κ or IgM λ and adjusted by the addition of purified IgM. The low pools for IgM κ and IgM λ was prepared from a normal serum sample adjusted with the addition of IgM depleted serum. A dilution series was prepared for IgM κ and IgM λ separately by combining the respective high and low pools to create a single proportional dilution series (e.g., 90+10, 80+20, etc.), to produce a total of 12 concentrations that covered the measuring range of the assays. Three replicates of each level of the dilution series were run and the mean calculated. Linearity was evaluated by calculating the percentage recovery at each concentration in the dilution series, and the %CV of the 3 replicates.

All of the concentrations within the analytical measuring range met the sponsor’s acceptance criterion of %CV < 8% and all concentrations met the sponsor’s

regression analysis acceptance criteria.

Linearity was demonstrated at the concentrations spanning the claimed measuring range. The observed values were graphed against the calculated values and a linear regression was performed. The regression plot equations where y is the measured level of IgMκ or λ and x is the theoretical concentration were:

Sample	Dilution Range (g/L)	Weighted Regression equation	Slope (95% CI)	Y-Intercept (95% CI)	R ²
IgMκ Sample	0.158 – 5.911	$y = 1.00x + 0.02$	1.00 (0.97 to 1.03)	0.02 (-0.08 to 0.12)	0.999
IgMλ Sample	0.131 – 5.085	$y = 0.97x + 0.02$	0.97 (0.96 to 0.99)	0.02 (-0.03 to 0.07)	1.00

The approximate measuring range of the Hevylite Human IgM Kappa kit for use on the SPA_{PLUS} is 0.2 – 5.0 g/L.

The approximate measuring range of the Hevylite Human IgM Lambda kit for use on the SPA_{PLUS} is 0.18 – 4.50 g/L.

Kit	Assay reportable range
Hevylite Human IgM Kappa kit for use on the SPA _{PLUS}	0.2 – 5.0 g/L
Hevylite Human IgM Lambda kit for use on the SPA _{PLUS}	0.18 – 4.50g/L

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

Traceability:

The calibrators, the master calibrator and controls are traceable to ERM-DA470k International Reference Material. The master calibrator is prepared from pooled human sera and is used to control calibration between lots.

Kit Stability:

The sponsor provided data to support the following stability claims of the Hevylite IgM Kappa and Hevylite IgM Lambda kits:

- Open-vial stability: Open kits are stable for up to 2 months when stored at 2 – 8°C.
- Real-time/Shelf-life stability: Unopened kits are stable for 12 months when stored at 2 – 8°C.
- On-board stability: Kits are stable for up to 35 days when stored on-board the instrument provided the temperature is maintained at 8 – 12°C.

d. *Detection limit:*

Analytical sensitivity was determined based on the recommendations contained in EP17-A Protocols for Determination of Limits of Detection and Limits of Quantitation, Approved Guideline.

Limit of Blank (LoB): The LoB is defined as the highest result expected in a sample that contains no analyte based on the 95th percentile distribution of blank results.

The LoB sample consisted of an analyte-depleted sample pool. The LoB sample was tested 60 times and the mean and standard deviation (SD) were calculated.

LoB was calculated using non-parametric analysis. The LoB results were ranked from lowest to highest concentration, and the LoB was estimated to be the result equivalent to the concentration at the 95th percentile, or between sample results 57 and 58. The LoB was determined to be 0.0 g/L for both the Hevylite Human IgM Kappa kit and the Hevylite Human IgM Lambda kit.

Limit of Detection (LoD): The LoD is defined as the lowest amount of analyte in a sample that can be reliably detected.

The LoD was calculated from the LoB and the combined SDs of the five LoQ samples: $LoD = LoB + [(1.645 \times SDs)]$. The LoD was determined to be 0.00115 g/L for the Hevylite Human IgM Kappa kit and 0.001645 g/L for the Hevylite Human IgM Lambda kit.

Limit of Quantitation (LoQ): The LoQ is defined as the lowest concentration at which the analyte can be quantified within predefined goals for bias and imprecision which are summarized in the following table:

Assay	Acceptance Criterion	Result
Hevylite Human IgM Kappa kit for use on the SPA _{PLUS}	LoD < LoQ	0.00115 g/L < 0.020 g/L
	TE < 0.019 g/L	0.0068 g/L
Hevylite Human IgM Lambda kit for use on the SPA _{PLUS}	LoD < LoQ	0.001645 g/L < 0.018 g/L
	TE < 0.012 g/L	0.0014 g/L

To determine LoQ for each kit, five normal donor samples with known IgM κ and IgM λ concentrations were diluted with analyte-depleted serum so that the concentrations were close to the bottom of the measuring range (IgM κ : 0.02 g/L and IgM λ : 0.018 g/L). The five samples for each kit were run 12 times over five days using the Hevylite Human IgM Kappa kit for use on the SPA_{PLUS} analyzer or the Hevylite Human IgM Lambda kit for use on the SPA_{PLUS}. The samples were also tested in triplicate over the same time period on the respective predicate device (BN II analyzer reagents). LoQ was calculated according to the formula $TE = |Bias| + 2 \times SDs$. The LoQ was determined to be 0.20 g/L for the Hevylite Human IgM Kappa kit

and 0.018 g/L for the Hevylite Human IgM Lambda kit.

Claimed Analytical sensitivity for each kit is summarized below:

Assay	LoB	LoD	LoQ
Hevylite Human IgM Kappa kit for use on the SPA _{PLUS}	0.0 g/L	0.0012 g/L	0.20 g/L
Hevylite Human IgM Lambda kit for use on the SPA _{PLUS}	0.0 g/L	0.0016 g/L	0.018 g/L

e. *Analytical specificity:*

Interfering Substances

This study was performed according to the recommendations contained in CLSI EP07-A2 Interference Testing in Clinical Chemistry, Approved Guideline, except that the cut-off for acceptable interference was set as $D_{obs} < D_{max}$ and $< \pm 10\%$ difference between the mean of the spiked sample and the unspiked sample. Twenty replicates of each of the serum base pools were run in order to calculate standard deviation. The D_{max} was set at 10% of the analyte concentration for each of the base pools. The standard deviation and the D_{max} were then used to calculate the number of replicates required in the interference study. Three replicates of each spiked and unspiked concentration were determined sufficient for this study.

Five (5) human serum-base pools were prepared to target five analyte concentrations spanning the measuring range. These levels were chosen to evaluate the effects the interferents at critical clinical concentrations (level 1 = within reference range; level 2= close to lower medical decision point; level 3 = close to upper medical decision point; level 4 = low clinical; level 5 = high clinical). The high concentration pool was diluted with analyte-free serum to achieve the desired target concentrations.

The serum pools were spiked separately with hemoglobin (5 g/L), bilirubin (200 mg/L), Intralipid (500 mg/dL) and triglyceride (1000 mg/dL). The spiked pools were analyzed, as were the unspiked pools, which were prepared by spiking the base pools separately with the same volume of commercially obtained blank for each potential interferent.

The results demonstrate that no significant interference was observed with the following interferents at the indicated concentrations tested:

Substance	Concentration tested
Bilirubin	200 mg/dL
Hemoglobin	5 g/L
Intralipid	500 mg/dL
Triglyceride	1000 mg/dL

Rheumatoid factor (RF) was not evaluated because this autoantibody is directed against the Fc portion of immunoglobulins. Interference is unlikely to take place in

the Hevylite Human IgM kappa and IgM Lambda assays, as the Fc region of the anti-IgM κ and anti-IgM λ antibodies are cleaved prior to coating onto the latex bead.

Cross-reactivity:

Cross-reactivity studies were carried out by testing Hevylite IgM Kappa and Hevylite IgM Lambda assays in the presence of high concentrations of potentially cross-reacting monoclonal proteins in 253 samples from a multiple myeloma library that included IgA1 κ , IgA1 λ , IgA2 κ , IgA2 λ , IgG1 κ , IgG1 λ , IgG2 κ , IgG2 λ , IgG3 κ , IgG3 λ , IgG4 κ , IgG4 λ , κ free light chain and λ free light chain multiple myeloma patient sera samples.

The samples were all tested for total IgG, total IgA, total IgM, and also with the Hevylite IgM Kappa and IgM Lambda assays. The results for total IgM were compared with the results obtained by the Hevylite IgM Kappa and IgM Lambda assays.

In addition, IgM κ patient samples were tested on Hevylite IgM Lambda kits to investigate potential cross-reactivity, and similarly, IgM λ patient samples were tested on IgM kappa kits.

No significant cross-reactivity was observed.

Antigen excess effect:

Prozone/Hook effect parameters are in effect to protect the SPA_{PLUS} analyzer from antigen excess effects. Reaction kinetics of high level samples was compared to that of the top calibrator for each kit. Samples detected as being in excess are flagged with a “P” flag.

The prozone flag limit for Hevylite IgM Kappa was set by testing 25 high level samples. The ‘P’ flag was then validated by analyzing 147 normal samples, 24 IgM κ library samples and 20 IgM κ myeloma quality control samples.

The prozone flag limit for Hevylite IgM Lambda was set by testing 24 high level samples. The ‘P’ flag was then validated by analyzing 147 normal samples, 20 IgM λ library samples and 10 IgM λ myeloma quality control samples.

f. Assay cut-off:

The cut-off values are the reference ranges for the normal population which have been established from the reference range study.

2. Comparison studies:

a. Method comparison with predicate device:

A total of 227 and 269 sera samples spanning the measuring range were assayed in singlicate on both Hevylite Human IgM kappa and IgM Lambda Kits for use on SPA_{PLUS} respectively, these results were compared to those obtained on the Hevylite Human IgM Kappa and IgM Lambda Kits for use on Siemens BNII.

The serum samples included 41 Waldenström’s Macroglobuliaemia patients, 23 normal donors and samples with elevated IgM κ and IgM λ levels.

Passing and Bablok regression are based on the balance of the paired results:

Assay Kit	N	Sample Range (g/L)	Regression Equation	Slope (95% CI)	Intercept (95% CI)
IgM Kappa	227	0.05–133.0	$y=0.84x + 0.07$	0.80 to 0.87	0.03 to 0.12
IgM Lambda	269	0.019–51.4	$y=0.93x + 0.0$	0.92 to 0.95	0.00 to 0.01
IgM κ /IgM λ ratio	222	0.001–2660	$y= 0.89x + 0.01$	0.88 to 0.91	0.00 to 0.03

Differences in instrument platforms may account for bias observed in the method comparison. The Hevylite™ Human IgM Kappa and Lambda Kits for use on SPA_{PLUS} have different (slightly lower) reference intervals than the predicate assays resulting in slightly higher results for the predicate. The SPA_{PLUS} instrument also uptakes a larger starting volume than the predicate which can result in aggregation; however kits have been optimized for optimal performance on each instrument platform. Limitations are included in the package inserts advising customers to establish reference ranges when possible.

In addition to linear regression, agreement between the Hevylite™ Human IgM Kappa and Lambda Kits for use on SPA_{PLUS} and predicate were determined. When considering the upper and the lower limits of the reference range as medical decision points for each assay and the ratios, the positive (abnormal result are outside reference range) and negative (normal result fall within reference range) agreement in the serum samples for each assay are presented in the tables below:

IgM Kappa		Predicate (normal = 0.29 – 1.82 g/L)		
		Positive	Negative	Total
Hevylite™ Human IgM Kappa Kit for use on SPAPLUS® (normal = 0.19 – 1.63 g/L)	Positive	159	2	161
	Negative	11	55	66
	Total	170	57	227

Positive percent agreement: 93.5% (95% CI: 88.7 to 96.7%)

Negative percent agreement: 96.5% (95% CI: 87.9 to 99.5%)

Overall agreement: 94.3% (95% CI: 88.3 to 95.6%)

IgM Lambda		Predicate (normal = 0.17 – 0.94 g/L)		
		Positive	Negative	Total
Hevylite™ Human IgM Lambda Kit for use on SPA _{PLUS} ® (normal = 0.12 – 1.01 g/L)	Positive	131	2	133
	Negative	16	120	136
	Total	147	122	269

Positive percent agreement: 89.1% (95% CI: 82.9 to 93.7%)

Negative percent agreement: 98.4% (95% CI: 94.2 to 99.8%)

Overall agreement: 93.3% (95% CI: 89.6 to 96.0%)

IgM κ /IgM λ Ratio		Predicate (normal = 0.17 – 2.3 g/L)		
		Positive	Negative	Total
Hevylite™ Human IgM Kappa and Lambda Kit for use on SPA _{PLUS} ® (normal = 1.18 – 2.74g/L)	Positive	164	14	478
	Negative	4	40	44
	Total	168	54	222

Positive percentage agreement: 97.6% (95% CI: 94.0 to 99.3%)

Negative percentage agreement: 74.1% (95% CI: 60.3 to 85.0%)

Overall agreement: 91.9% (95% CI: 85.2 to 93.5%)

b. *Matrix comparison:*

Not applicable.

3. Clinical studies:

a. *Clinical Sensitivity and Specificity:*

Not applicable.

b. Other clinical supportive data (when a. is not applicable):

Not applicable.

4. Clinical cut-off:

Not applicable.

5. Expected values/Reference range:

The normal ranges established in accordance with CLSI C28-A3 using 147 UK adult blood donors (27% male, 36% female, and 37% unknown). The assays were performed on the SPA_{PLUS} analyzer. A non-parametric analysis of specimens for the distribution of IgM κ and IgM λ was performed.

Test Parameter	IgM κ	IgM λ	IgM κ/λ ratio
N	147		
Range	0.154 – 2.18 g/L	0.091 – 1.321 g/L	0.901 – 3.832 g/L
Mean	0.708 g/L	0.394 g/L	1.845 g/L
Median	0.627 g/L	0.346 g/L	1.814 g/L
95 th percentile	0.193 – 1.633 g/L	0.120 – 1.013 g/L	1.182 – 2.735 g/L
90% CI lower boundary	0.154 – 0.253	0.091 – 0.151	0.901 – 1.326
90% CI upper boundary	1.361–2.183	0.7650 – 1.321	2.483 – 3.832

The upper and lower limits of the reference range for IgM κ (0.19 – 1.63 g/L), IgM λ (0.12 – 1.01 g/L) and the IgM κ / λ ratio (1.18 – 2.74 g/L) are defined as the “cut-offs”. Samples with a Hevylite result above any of these reference ranges, or below the lower cut-off are classified as abnormal.

Normal range results as included in IFU:

Normal adult serum	Mean	Median	95 Percentile Range
IgM kappa (g/L)	0.71	0.63	0.19 – 1.63
IgM lambda (g/L)	0.39	0.35	0.12 – 1.01
IgM κ / IgM λ ratio (g/L)	1.85	1.81	1.18 – 2.74

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.