

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

**A. 510(k) Number:**

K161854

**B. Purpose for Submission:**

New instrument for a cleared IVD assay

**C. Measurand:**

Immunoglobulin IgG Kappa (combined  $\gamma$  heavy and  $\kappa$  light chain) and Immunoglobulin IgG Lambda (combined  $\gamma$  heavy and  $\lambda$  light chain)

**D. Type of Test:**

Quantitative, Turbidimetry

**E. Applicant:**

The Binding Site Group, Ltd.

**F. Proprietary and Established Names:**

Hevylite<sup>®</sup> Human IgG Kappa Kit for use on the SPAPLUS<sup>®</sup>  
Hevylite<sup>®</sup> Human IgG Lambda Kit for use on the SPAPLUS<sup>®</sup>

**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:

PCN - IgG Kappa (Heavy and Light chain Combined). Antigen, antiserum, control  
PCO - IgG Lambda (Heavy and Light chain Combined). Antigen, antiserum, control

4. Panel:

Immunology (82)

**H. Intended Use:**

1. Intended use(s):

Hevylite Human IgG Kappa is a quantitative in vitro assay performed on the SPAPLUS<sup>®</sup> analyzer for the measurement of IgG Kappa (IgG heavy chain and kappa light chain intact immunoglobulin) in serum. Measurement of Hevylite Human IgG Kappa is used alongside Hevylite Human IgG Lambda to calculate the IgG Kappa/IgG Lambda ratio. The Hevylite Human IgG Kappa/IgG Lambda ratio can be used when monitoring previously diagnosed IgG multiple myeloma and is used in conjunction with other laboratory tests and clinical evaluations. The assignment of complete response is reliant upon other tests including immunofixation, bone marrow and urine assessments.

Hevylite Human IgG Lambda is a quantitative in vitro assay performed on the SPAPLUS<sup>®</sup> analyzer for the measurement of IgG Lambda (IgG heavy chain and lambda light chain intact immunoglobulin) in serum. Measurement of Hevylite Human IgG Lambda is used alongside Hevylite Human IgG Kappa to calculate the IgG Kappa/IgG Lambda ratio. The Hevylite Human IgG Kappa/IgG Lambda ratio can be used when monitoring previously diagnosed IgG multiple myeloma and is used in conjunction with other laboratory tests and clinical evaluations. The assignment of complete response is reliant upon other tests including immunofixation, bone marrow and urine assessments.

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 IgG Kappa in a given specimen determined with assays with different manufacturers or different instrument platforms 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 IgG Kappa assay used. Values obtained with different assay methods cannot be used interchangeably. If, in the course of serially monitoring a patient, the assay method used for determining Hevylite IgG Kappa levels is changed, additional sequential testing should be carried out. Prior to changing assays, the laboratory MUST confirm baseline values for patients being serially monitored.

Warning: The result of Hevylite Human IgG Lambda in a given specimen determined with assays with different manufacturers or different instrument platforms 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 IgG Lambda assay used. Values obtained with different assay methods cannot be used interchangeably. If, in the course of serially monitoring a patient, the assay method used for determining Hevylite Human IgG Lambda levels is changed, additional sequential testing should be carried out. Prior to changing assays, the laboratory MUST confirm baseline values for patients being serially monitored.

4. Special instrument requirements:

The Binding Site SPAPLUS<sup>®</sup> analyzer

**I. Device Description:**

The Hevylite Human IgG Kappa Antiserum and Hevylite Human IgG Lambda Antiserum contain vials of ready-to-use polyclonal monospecific sheep anti-IgG antisera against combined  $\gamma$  heavy and  $\kappa$  light chain or combined  $\gamma$  heavy and  $\lambda$  light chain, calibrators (six levels), controls (low and high) and reaction buffer in liquid form. The reagents contain 0.099% sodium azide, 0.1% E-amino-n-caproic acid (EACA), 1mM ethylenediaminetetraacetic acid (EDTA) and 0.01% benzamidine as preservatives.

**J. Substantial Equivalence Information:**

1. Predicate device names and predicate 510(k) number:

Hevylite<sup>®</sup> Human IgG Kappa Kit and Hevylite<sup>®</sup> Human IgG Lambda Kit for use on Siemens BN<sup>™</sup> II Systems (K132555)

2. Comparison with predicate:

Similarities		
Item	Device Hevylite <sup>®</sup> IgG Kappa (IgG $\kappa$ ) and IgG Lambda (IgG $\lambda$ ) Kits	Predicate Hevylite <sup>®</sup> IgG Kappa (IgG $\kappa$ ) and IgG Lambda (IgG $\lambda$ ) Kits
Intended Use	Quantitative in vitro assay for the measurement of IgG Kappa (IgG heavy chain and Kappa light chain intact immunoglobulin) and IgG Lambda (IgG heavy chain and Lambda light chain intact immunoglobulin) in serum. Measurement of Hevylite Human IgG Kappa is used alongside Hevylite	Same

<b>Similarities</b>		
<b>Item</b>	<b>Device</b>	<b>Predicate</b>
	Hevylite <sup>®</sup> IgG Kappa (IgGκ) and IgG Lambda (IgGλ) Kits	Hevylite <sup>®</sup> IgG Kappa (IgGκ) and IgG Lambda (IgGλ) Kits
	Human IgG Lambda or the measurement Hevylite Human IgG Lambda is used alongside Hevylite Human IgG Kappa to calculate the IgG Kappa/IgG Lambda ratio. The Hevylite Human IgG Kappa/IgG Lambda ratio can be used when monitoring previously diagnosed IgG multiple myeloma and is used in conjunction with other laboratory tests and clinical evaluations. The assignment of complete response is reliant upon other tests including immunofixation, bone marrow and urine assessments.	
Analyte	IgG Kappa and IgG Lambda	Same
Antibody	Polyclonal monospecific Sheep anti-human combined γ heavy and κ light chain or combined γ heavy and λ light chain	Same
Control	Binding Site High and Low Controls	Same
Traceability	ERM-DA470k/IFCC	Same
Sample Matrix	Serum	Same

<b>Differences</b>		
<b>Item</b>	<b>Device</b>	<b>Predicate</b>
Instrument	The Binding Site SPAPLUS <sup>®</sup>	Siemens BN <sup>™</sup> II Systems
Method	Turbidimetry	Nephelometry
Calibrator	Six levels of Binding Site Hevylite Calibrator	One level of Binding Site Hevylite Calibrator auto-diluted to six different concentrations
On-board Stability	1 month	Not stated
Open Vial Stability	2 months	3 months
Measuring Range	At standard 1/20 dilution:	At standard 1/100 dilution:

Differences		
Item	Device	Predicate
	IgGκ: 1.9–40.0 g/L IgGλ: 0.92–29.5 g/L  Extended Range for IgGκ: 1/1 dilution: 0.09–2.0 g/L 1/80 dilution: 7.5–160.0 g/L  Extended Range for IgGλ: 1/1 dilution: 0.05–1.48 g/L 1/80 dilution: 3.68–118.0 g/L	IgGκ: 1.72–27.5 g/L IgGλ: 0.88–14.0 g/L  Extended Range for IgGκ: 1/5 dilution: 0.09–1.38 g/L 1/20 dilution: 0.34–5.50 g/L 1/400 dilution: 6.88–110.0 g/L 1/2000 dilution: 34.4–550 g/L  Extended Range for IgGλ: 1/5 dilution: 0.04–0.70 g/L 1/20 dilution: 0.18–2.80 g/L 1/100 dilution: 0.88–14.0 g/L 1/400 dilution: 3.50–56.0 g/L 1/2000 dilution: 17.5–280 g/L
Reference Interval	IgGκ: 3.84–12.07 g/L IgGλ: 1.91–6.74 g/L IgGκ/IgGλ Ratio: 1.12–3.21	IgGκ: 4.03–9.78 g/L IgGλ: 1.97–5.71 g/L IgGκ/IgGλ Ratio: 0.98–2.75

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

- CLSI EP05-A: Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline–Second Edition
- CLSI EP6-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach
- CLSI EP07-A2: Interference Testing in Clinical Chemistry; Approved Guideline–Second Edition
- CLSI EP17-A: Protocols for Determination of Limits of Detection and Limits of Quantitation; Approved Guideline
- CLSI C28-A3: Defining, Establishing and Verifying Reference Intervals in the Clinical Laboratory

**L. Test Principle:**

Evaluating the concentration of a soluble antigen (e.g. IgG Lambda) by turbidimetry involves the addition of the test sample to a solution containing the appropriate antibody (anti-IgG Lambda) 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:

All of the manufacturer’s pre-specified acceptance criteria were met.

a. *Precision/Reproducibility:*

The study design followed the recommendations of CLSI EP05-A2. The within-run, between-run, between-day, between-lot and between-instrument precision were determined by testing three IgGκ and three IgGλ serum samples over 21 days with two runs per day and two replicates per run on three different reagent lots on three analyzers for IgGκ and three different reagent lots on three analyser for IgGλ. Results are summarized below.

IgGκ precision studies:

Sample	Mean (g/L)	Within Run		Between Run		Between Day		Between Lot		Between Instrument		Total	
		SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	25.18	0.32	1.2	0.52	2.0	1.53	5.7	1.37	5.4	0.34	1.4	1.65	6.1
2	9.69	0.11	1.2	0.25	2.6	0.43	4.4	0.06	0.6	0.08	0.9	0.51	5.2
3	3.18	0.07	2.3	0.13	4.1	0.16	5.3	0.07	2.0	0.07	2.3	0.22	7.1

IgGλ precision studies:

Sample	Mean (g/L)	Within Run		Between Run		Between Day		Between Lot		Between Instrument		Total	
		SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	23.75	0.15	0.7	0.57	2.5	1.23	5.3	1.4	5.9	0.30	1.3	1.36	5.9
2	5.33	0.08	1.4	0.14	2.6	0.28	5.2	0.20	3.7	0.06	1.2	0.32	6.0
3	1.73	0.03	2.1	0.07	4.7	0.14	8.7	0.12	6.6	0.01	5.5	0.16	10.1

b. *Linearity/assay reportable range:*

A linearity study was performed following CLSI EP6-A. The linearity of the Hevylite IgG Kappa and Hevylite IgG Lambda assays was confirmed using serially diluted serum samples (IgGκ myeloma sample diluted with IgGκ depleted sample and IgGλ elevated polyclonal sample diluted with IgGλ depleted sample) to cover the standard (1/20) dilution measuring range 1.9–40 g/L and 0.92–29.5 g/L, respectively. Deviation from linearity was ≤ 5.7% for IgGκ and ≤ 3.2% for IgGλ. The weighted linear regression observed versus % high pool was  $y = 48.72x + 0.29$  for IgGκ and  $y = 38.03x + 1.79$  for IgGλ.

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

Traceability:

An internal reference (IR) material was assigned by comparison with the Reference Material ERM-DA470k/IFCC.

Stability:

A real-time stability study of unopened kits was performed on three lots of Hevylite IgG Kappa and Hevylite IgG Lambda SPAPLUS® kits with testing time intervals at 0, 1, 3, 6, 10, and 13 months. Data support a shelf life claim of 12 months at 2–8°C.

Open-vial stability was performed on three lots of Hevylite IgG Kappa and Hevylite IgG Lambda SPAPLUS® kits with testing time intervals at 0, 28, and 84 days. Data support the open vial stability claim of 2 months at 2–8°C.

On-board stability was performed on three lots of Hevylite IgG Kappa and Hevylite IgG Lambda SPAPLUS® kits with testing time intervals at 0, 7, 14, 21, 28 and 35 days. Data support the on-board stability claim of 1 month, provided that the power is left switched on as stated in the product insert.

d. *Detection limit:*

The analytical sensitivity was determined according to CLSI EP17-A. The Limits of Blank (LoB) were based on 60 determinations of analyte-depleted sample and was estimated at the 95<sup>th</sup> percentile of the distribution. The Limits of Detection (LoD) were calculated using the formula  $LoD = LoB + c\beta SDs$ , where  $c\beta$  is 1.645 and SD is the combined standard deviation of measurements of five low level serum samples diluted with analyte depleted serum to achieve a concentration close to the bottom of the measuring range. The samples were tested twelve times over five days and the total errors (0.0184 g/L for IgG $\kappa$  and 0.0060 g/L for IgG $\lambda$ ) were within the maximum allowable total error. The Limits of Quantitation (LoQ) were calculated from the total error of the LoD study. The tabulated summary is shown below:

	LoB	LoD	LoQ
Hevylite IgG Kappa	0.000 g/L	0.008 g/L	0.095 g/L
Hevylite IgG Lambda	0.000 g/L	0.004 g/L	0.046 g/L

e. *Analytical specificity:*

Interference:

Interferences were assessed according to CLSI EP07-A2 by testing three serum samples with different IgG $\kappa$  and IgG $\lambda$  concentration ranges: (1) within the reference interval, IgG $\kappa$  6.12 and 7.96 g/L and IgG $\lambda$  4.54 g/L; (2) close to the medical decision point, IgG $\kappa$  11.78 and 12.45 g/L and IgG $\lambda$  6.63 g/L; (3) above the medical decision point, IgG $\kappa$  18.23 and 21.22g/L and IgG $\lambda$  14.3 g/L. Each sample was spiked with interfering substances and tested in duplicate.

No significant assay interference effects were observed when the three samples of IgG $\kappa$  and IgG $\lambda$  were tested with bilirubin at 200 mg/L, hemoglobin at 5 g/L, triglyceride at 1000 mg/dL or Intralipid at 2000 mg/dL (IgG $\kappa$ ) or 250 mg/dL (IgG $\lambda$ ) or the 16 commonly used drugs at the concentrations given below.

Drug	Concentration tested
Acetaminophen	1324 $\mu$ mol/L
Acetylsalicylic Acid	3.63 mmol/L
Ascorbic Acid	342 $\mu$ mol/L
Bortezomib	6 mg/mL
Caffeine	308 $\mu$ mol/L
Cimitidine	79.2 $\mu$ mol/mL
Cyclophosphamide Monohydrate	60 $\mu$ g/L
Digoxin	7.8 nmol/L
Furosemide	90 $\mu$ mol/L
Ibuprofen	2425 $\mu$ mol/L
Methotrexate	2 mmol/L
Penicillin	75 mg/L
Phenytoin	198 $\mu$ mol/L
Pomalidomide	100 $\mu$ g/mL
Prednisolone	100 $\mu$ g/mL
Theophylline	222 $\mu$ mol/L

Cross-reactivity:

For IgG $\kappa$ , the possibility of cross-reactivity with other immunoglobulin heavy/light chain combinations was assessed by testing eight IgG $\lambda$ , seven IgA $\kappa$ , eight IgA $\lambda$ , 14 IgM $\kappa$ , eight IgM $\lambda$ , two Free Kappa and one Free Lambda myeloma samples. No significant cross-reactivity was observed.

For IgG $\lambda$ , the possibility of cross-reactivity with other immunoglobulin heavy/light chain combinations was assessed by testing 13 IgG $\kappa$ , seven IgA $\kappa$ , eight IgA $\lambda$ , 14 IgM $\kappa$ , eight IgM $\lambda$ , two Free Kappa and one Free Lambda myeloma samples. No significant cross-reactivity was observed.

Antigen Excess:

The possibility of antigen excess occurring when using the device on The Binding Site SPAPLUS<sup>®</sup> was evaluated with 13 monoclonal IgG $\kappa$  and 15 monoclonal IgG $\lambda$  samples with concentrations above the 1.9–40.0 g/L and 0.92–29.5 g/L standard measuring ranges, respectively. No antigen excess effect up to 109.7 g/L of IgG $\kappa$  and 108.4 g/L of IgG $\lambda$  was observed at 1 + 9 dilution except for one IgG $\kappa$  sample. If antigen excess is suspected, the P.I. Limitation section states as follows:

*“Monoclonal immunoglobulins are highly variable. Any sample giving unexpected results should be retested at a higher dilution (lower concentration) to preclude antigen excess”.*

f. Assay cut-off:

Refer to Expected values

2. Comparison studies:

a. Method comparison with predicate device:

IgG Kappa:

A comparison study was performed by analysing 217 samples, including 91 IgG Kappa paraprotein and 48 IgG Lambda paraprotein samples, 41 donor samples and 37 other clinical samples, covering the range 0.2–62.7 g/L, using the SPAPLUS® Hevylite IgG Kappa Kit and the predicate device. Passing Bablok regression analysis generated the results below.

IgG Lambda:

A comparison study was performed by analysing 171 samples, including 64 IgG Kappa paraprotein and 66 IgG Lambda paraprotein samples and 41 donor samples covering the range 0.06–43.96 g/L, using the SPAPLUS® Hevylite IgG Lambda Kit and the predicate device. Passing Bablok regression analysis generated the results below.

IgG Kappa/ IgG Lambda Ratio

A comparison study was performed by analysing 115 samples, including 38 IgG Kappa paraprotein and 36 IgG Lambda paraprotein samples and 41 donor samples, covering the range 0.03–488.66 g/L using the SPAPLUS® Hevylite IgG Lambda Kit and the predicate device. Passing Bablok regression analysis generated the results below.

	N	Sample Range	Passing & Bablok	Slope 95% CI	Intercept 95% CI
IgG Kappa	219	0.2–62.7 g/L	$y = 1.15x - 0.51$	1.09 to 1.22	-0.98 to 0.19
IgG Lambda	117	0.06–43.96 g/L	$y = 1.02x + 0.24$	0.95 to 1.06	0.05 to 0.53
IgGκ/IgGλ ratio	115	0.03–488.66 g/L	$y = 0.99x - 0.07$	0.91 to 1.13	-0.26 to 0.04

b. Matrix comparison:

Not applicable

3. Clinical studies:

a. Clinical Sensitivity and Clinical Specificity:

Not applicable

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

Data modeling of the BN II Study onto SPAPLUS® Study

Purpose of the study:

The purpose of this study was to compare the clinical Hevylite Chain (HLC) Response categorization of the predicate BN II results and results from the Hevylite IgG Kappa and Hevylite IgG Lambda Kits for use on SPAPLUS®. The BN II results were obtained from samples taken at multiple time points from IgG Kappa and IgG Lambda multiple myeloma patients during the course of their disease. The Sponsor generated Passing and Bablok regression equations for the comparison study of the SPAPLUS® kits against the predicate kits. The regression equation was then used to model a response from the existing 437 monitoring sample results from the original BNII submission to evaluate the clinical validity of the new device.

<b>HLC Monitoring Response Categories</b>	
Complete Response (CR)	HLC ratio within the normal range, negative urine immunofixation.
Very Good Partial Response (VGPR)	> 91% reduction of HLC ratio from baseline and urine M protein level < 100 mg/24 hrs.
Partial Response (PR)	Reduction of HLC ratio from baseline between 47–91% and reduction in 24 hr urinary M-protein by $\geq 90\%$ or $\leq 200$ mg/24 hrs.
Stable Disease (SD)	A change in HLC ratio from baseline < 32% increase but < 47% reduction
Progressive Disease result(PD)	$\geq 32\%$ increase in HLC ratio from baseline (the absolute increase in involved IgG must be $\geq 5$ g/L) or $\geq 25\%$ increase in urine M-component from baseline (the absolute increase must be $\geq 200$ mg/24 hrs).

HLC Response Category Study using SPAPLUS®

Assignment of classification was based on the HLC Monitoring Response Category detailed in Table 1, using all assay data available. Responses were categorized in accordance with the modified NCCN Guidelines v.1.2011 by using the percentage (%) change in Serum Protein Electrophoresis (SPEP) or total IgG from baseline. Responses were characterized as progressive disease (PD), stable disease (SD), partial response (PR), very good partial response (VGPR) and complete response (CR). Kappa Statistics was used to evaluate the agreement between test and the clinical status as determined by clinical evaluation combined with either the test or predicate device.

Monitoring Study using SPAPLUS®

A comparison study using 63 monitoring samples from 21 patients (11 IgG Kappa and 10 IgG Lambda) was performed to compare the BN II HLC response categories assigned to those observed with the SPAPLUS® Hevylite IgG Kappa and IgG Lambda Kits. (Note: The SPAPLUS® HLC monitoring response categories are modified from the NCCN v1.2011 Treatment Response Classification categories. The SPAPLUS®

HLC Monitoring Response Category is not to be used interchangeably with other manufacturer’s assays or with any other instrument platform monitoring response category.) The results of comparison study using 63 monitoring samples yielding 42 response classifications are shown in the table below:

Observed		BN II HLC* Response Category					
		CR*	VGPR	PR	SD	PD	Total
SPAPLUS® HLC Response Category	CR**	7	0	1	0	0	8
	VGPR	1	1	0	0	0	2
	PR	0	0	1	1	0	2
	SD	0	0	0	29	1	30
	PD	0	0	0	0	0	0
	Total	8	1	2	30	1	42
Weighted Kappa (95% CIs)		0.94 (0.87–1.00)					
Kappa (95% CIs)		0.79 (0.61–0.97)					
*HLC: HevyLite Chain							
** The assignment of complete response (CR) is reliant upon other tests including immunofixation, bone marrow and urine assessments							

These monitoring data were also supported by additional statistical regression equation modelling data.

#### Data Modeling Study

Data modeling was carried out with patient samples that were not included in the SPAPLUS® Response Category Study. The Passing and Bablok regression equation derived from the previously cleared 437 BN II data set were mathematically transformed using the SPAPLUS® regression equation. The statistical regression equation modelling (Cohen’s Kappa) results are summarized in the tables below.

Observed		BN II Assigned HLC* Response Category					
		CR	VGPR	PR	SD	PD	Total
SPAPLUS® H/L** transformed data	CR	51	3	1	0	0	55
	VGPR	6	69	17	0	0	92
	PR	3	10	175	8	0	196
	SD	0	0	5	88	0	93
	PD	0	0	0	0	1	1
	Total	60	82	198	96	1	437
Weighted Kappa (95% CIs)		0.92 (95% CIs: 0.89–0.94)					
Kappa (95% CIs)		0.83 (95% CIs: 0.78–0.87)					
*HLC: HevyLite Chain							
**H/L: For H/L - applied the imprecision values to generate the highest (H) Kappa and the lowest (L) Lambda results possible.							

Observed		BN II Assigned HLC* Response Category					
		CR	VGPR	PR	SD	PD	Total
SPAPLUS®  L/H** transformed data	CR	47	28	22	1	0	98
	VGPR	12	44	12	0	0	68
	PR	1	10	159	7	0	177
	SD	0	0	5	88	0	93
	PD	0	0	0	0	1	1
	Total	60	82	198	96	1	437
Weighted Kappa (95% CIs)		0.81 (95% CI: 0.76–0.85)					
Kappa (95% CIs)		0.68 (95% CI: 0.63–0.74)					
*HLC: HevyLite Chain ** L/H: For L/H - applied the imprecision values to generate the lowest (L)Kappa and the highest (H) Lambda results possible.							

The SPAPLUS® IgG HLC Monitoring Response Category in the two Tables above were modified from the NCCN v1.2011 *Guidelines on Treatment Response Classification* as shown in the Table below.

Response	NCCN v1.2011	Disease Monitoring Using HLC SPAPLUS® IgG
Complete Response (CR)	<b>Negative IFE</b> on the serum and urine and disappearance of any soft tissue plasmacytomas and ≤ 5% plasma cells in bone marrow	<b>HLC ratio</b> within the <b>normal range</b> (SPAPLUS® IgGκ/IgGλ Ratio 1.12–3.21) and negative urine immunofixation
Very Good Partial Response (VGPR)	Serum and urine <b>M protein detectable by IFE but not SPEP</b> or ≥ 90% reduction in serum M protein level plus urine M protein level < 100 mg per 24 hours	> 91% reduction of HLC ratio from baseline and urine M protein level < 100 mg per 24 hours
Partial Response (PR)	≥ <b>50% reduction of serum M protein</b> and reduction in 24 hour urinary M protein by ≥ 90% or to < 200 mg per 24 hours.	<b>Reduction of HLC ratio from baseline between 47–91%</b> and reduction in 24 hour urinary M protein by ≥ 90% or to < 200 mg per 24 hours.
Stable Disease (SD)	Not meeting criteria for CR, VGPR, PR or progressive disease	A change in HLC ratio from baseline < <b>32% increase but &lt; 47% reduction.</b>

Response	NCCN v1.2011	Disease Monitoring Using HLC SPAPLUS® IgG
Progressive Disease (PD)	<p><b>Increase of <math>\geq 25\%</math></b> from baseline in 1 or more:</p> <ul style="list-style-type: none"> <li>• Serum M-component and/or (the absolute increase must be <math>\geq 0.5</math> g/dL)</li> <li>• Urine M component and/or (the absolute increase must be <math>\geq 200</math> mg per 24 hours)</li> <li>• Bone marrow plasma cell percentage: the absolute percentage must be <math>\geq 10\%</math></li> <li>• Definite development of new bone lesions or soft tissue plasmacytomas or definite increase in the size of existing bone lesions or soft tissue plasmacytomas</li> <li>• Development of hypercalcemia</li> </ul>	<p><b><math>\geq 32\%</math> increase in HLC ratio</b> from baseline (the absolute increase in involved IgG must be <math>\geq 5</math> g/L) or a <math>\geq 25\%</math> increase in urine M-component from baseline (the absolute increase must be <math>\geq 200</math> mg per 24 hours)</p>

4. Clinical cut-off:

Refer to discussion above.

5. Expected values/Reference range:

Reference Adult Serum	Mean	Median	95 <sup>th</sup> Percentile Range
IgG Kappa (g/L)	7.10	6.75	3.84–12.07
IgG Lambda (g/L)	3.95	3.90	1.91–6.74
IgG Kappa/IgG Lambda Ratio	1.84	1.74	1.12–3.21

**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.