

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

A. 510(k) Number:

K152389

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 Immunoturbidimetric

E. Applicant:

The Binding Site Group, Ltd.

F. Proprietary and Established Names:

Optilite® Hevylite® IgM Kappa Kit
Optilite® Hevylite® IgM Lambda Kit

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):

Optilite Hevylite IgM Kappa Kit:

The Optilite Hevylite IgM Kappa Kit is intended for the *in vitro* quantitative measurement of IgM kappa (combined μ heavy and κ light chain) in serum using the Binding Site Optilite analyser. The test result is to be used with previously diagnosed Waldenström's macroglobulinaemia. The test result should be used in conjunction with other laboratory and clinical findings.

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

Optilite Hevylite IgM Lambda Kit:

The Optilite Hevylite IgM Lambda Kit is intended for the *in vitro* quantitative measurement of IgM lambda (combined μ heavy and λ light chain) in serum using the Binding Site Optilite analyser. The test result is to be used with previously diagnosed Waldenström's macroglobulinaemia. The test result should be used in conjunction with other laboratory and clinical findings.

This assay has not been established for the diagnosis, monitoring and prognosis of Waldenström'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 IgM Kappa in a given specimen determined with assays with different manufacturers and different 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 IgM Kappa assay used. Values obtained with different assay methods cannot be used interchangeably.

Warning: The result of Hevylite IgM Lambda in a given specimen determined with assays with different manufacturers and different 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 IgM Lambda assay used. Values obtained with different assay methods cannot be used interchangeably.

4. Special instrument requirements:

Binding Site Optilite®

I. Device Description:

The Optilite® Hevylite® IgM Kappa Kit and Optilite® Hevylite® IgM Lambda Kit contain polystyrene latex coated with a polyclonal monospecific sheep anti-human IgM antibody against combined μ heavy and κ light chain or combined μ heavy and λ light chain; a

calibrator; two controls (low and high) and a reaction buffer. The reagents contain 0.099% sodium azide, 0.1% E-amino-n-caproic acid (EACA), 0.01% benzamidine and 0.05% ProClin™ as preservatives.

J. Substantial Equivalence Information:

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

Hevylite™ Human IgM Kappa Kit for use on SPA_{PLUS}, and Hevylite™ Human IgM Lambda Kit for use on SPA_{PLUS}, K140686

2. Comparison with predicate:

Similarities		
Item	Device Optilite Hevylite IgM Kappa and IgM Lambda kits	Predicate Hevylite IgM Kappa and IgM Lambda kits use on SPA _{PLUS}
Intended Use	Quantification of IgM κ (combined μ heavy and λ light chain) or IgM λ (combined μ heavy and λ light chain) concentration in human serum	Same
Indications for Use	Used with previously diagnosed Waldenström’s macroglobulinemia and in conjunction with other clinical and laboratory findings.	Same
Type of Test	Quantitative	Same
Detection Method	Turbidimetric	Same
Sample Matrix	Serum	Same
Controls	One low and one high control (serum, ready to use)	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
Reference Range	IgM κ : 0.19–1.63 g/L IgM λ : 0.12–1.01 g/L IgM κ/λ ratio: 1.18–2.74 g/L	Same
On-board Vial stability	1 month	Same

Differences		
Item	Device Optilite Hevylite IgM Kappa and IgM Lambda kits	Predicate Hevylite IgM Kappa and IgM Lambda kits use on SPA _{PLUS}
Instrument	Binding Site Optilite Analyser	Binding Site SPA _{PLUS} Analyser
Calibrator	One calibrator and Optilite analyser dilutes the calibrator on board	Set of six calibrators
Analyser dilution	No manual pre-dilution step required. The Optilite analyser can carry out a 1 + 299 (1/300) samples dilution	Manual pre-dilution step for a 1/250 sample dilution
Measuring range	IgMκ: 0.2–5.0 g/L IgMλ: 0.18–4.5 g/L (at standard 1/10 dilution) Extended Range for IgMκ: 1 + 0 dilution: 0.02–0.50 g/L 1 + 9 dilution: 0.2–5.0 g/L 1 + 89 dilution: 1.8–45 g/L 1 + 299 dilution: 6.0–150 g/L Extended Range for IgMλ: 1 + 0 dilution: 0.018–0.450 g/L 1 + 9 dilution: 0.18–4.50 g/L 1 + 89 dilution: 1.62–40.50 g/L 1 + 299 dilution: 5.4–135.0 g/L	IgMκ: 0.2–5.0 g/L IgMλ: 0.18–4.5 g/L (at standard 1/10 dilution) 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: 1.62–40.5 g/L 1/250 dilution: 4.5–112.5 g/L
Open Vial stability	3 months	1 month

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

CLSI EP05-A2: Evaluation of Precision Performance of Clinical Chemistry.

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

CLSI EP17-A: Determination of Limits of Detection and Limits of Quantitation.

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

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

L. Test Principle:

The determination of soluble antigen concentration by turbidimetric methods involves the reaction with specific antiserum to form insoluble complexes. When light is passed through the suspension formed, a portion of the light is transmitted and focused onto a photodiode by an optical lens system. The amount of transmitted light is indirectly proportional to the specific protein concentration in the test sample. Concentrations are automatically calculated by reference to a calibration curve stored within the instrument.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

The study was carried out based on EP05-A2 *Evaluation of Precision Performance of Quantitative Measurement Methods*. Eight sera samples with different concentrations of IgM Kappa or IgM Lambda that span the measuring range of the assays at 0.2–5.0 g/L and 0.18–4.50 g/L respectively. The eight samples were tested in 2 runs per day over 21 days using 3 reagent lots on 3 Optilite analysers. Results met the acceptance criteria for total precision ($\leq 10\%$ CV), within run precision ($\leq 5\%$ CV), between run precision ($\leq 8\%$ CV), and between day precision ($\leq 8\%$ CV). Results are summarized below:

Optilite Hevylite IgM Kappa Kit

IgMκ Sample	N	Mean g/L	Within-Run		Between-Run		Between-Day		Total Precision	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
1*	84	0.10	0.00	1	0.00	2	0.01	5.2	0.01	5.7
2*	84	0.15	0.00	1.1	0.00	2.5	0.01	5	0.01	5.7
3*	84	0.22	0.00	1.1	0.01	2.9	0.01	4.2	0.01	5.2
4	84	0.33	0.00	1	0.02	4.5	0.00	0	0.02	4.6
5	84	1.19	0.01	1.1	0.05	3.8	0.00	0	0.05	3.9
6	84	2.05	0.03	1.2	0.09	4.3	0.00	0	0.10	4.5
7	84	2.60	0.05	1.7	0.13	4.9	0.03	1	0.14	5.2
8	84	3.94	0.06	1.5	0.18	4.6	0.11	2.9	0.22	5.6

* performed at the 1 + 0 sample dilution

IgMκ Sample	N	Mean g/L	Between-Instrument		Between-lot	
			SD	%CV	SD	%CV
1*	84	0.10	0.00	0.5	0.00	3.9
2*	84	0.15	0.00	1.6	0.01	4.3
3*	84	0.22	0.00	1.4	0.01	3.2
4	84	0.33	0.00	0.8	0.00	1.3
5	84	1.19	0.01	0.9	0.02	1.3

IgMκ Sample	N	Mean g/L	Between- Instrument		Between-lot	
			SD	%CV	SD	%CV
6	84	2.05	0.03	1.5	0.03	1.6
7	84	2.60	0.05	1.9	0.06	2.3
8	84	3.94	0.09	2.2	0.09	2.3

* performed at the 1 + 0 sample dilution

Optilite Hevylite IgM Lambda Kit

IgMλ Sample	N	Mean g/L	Within-Run		Between-Run		Between-Day		Total Precision	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
1*	84	0.06	0.00	1.5	0.00	0.9	0.00	4.7	0.00	5
2*	84	0.09	0.00	1.2	0.00	1.4	0.00	3.9	0.00	4.3
3*	84	0.13	0.00	0.9	0.00	1.4	0.00	2.8	0.01	3.3
4*	84	0.36	0.00	1.1	0.01	1.6	0.01	2.9	0.01	3.5
5	84	0.76	0.01	1.5	0.01	1.7	0.02	2.3	0.03	3.2
6	84	1.22	0.01	1.1	0.02	1.5	0.03	2.5	0.04	3.2
7	84	1.46	0.01	0.8	0.02	1.4	0.05	3.3	0.05	3.7
8	84	3.90	0.06	1.4	0.04	1.1	0.10	2.7	0.13	3.2

* performed at the 1 + 0 sample dilution

IgMλ Sample	N	Mean g/L	Between- Instrument		Between-lot	
			SD	%CV	SD	%CV
1*	84	0.06	0.00	2.8	0.00	4.3
2*	84	0.09	0.00	3.4	0.00	2.5
3*	84	0.13	0.00	1.5	0.00	2
4*	84	0.36	0.00	0.7	0.01	3.2
5	84	0.76	0.00	0.3	0.01	1.7
6	84	1.22	0.01	0.9	0.02	1.7
7	84	1.46	0.01	0.5	0.03	1.9
8	84	3.90	0.08	2.1	0.02	0.6

* performed at the 1 + 0 sample dilution

b. Linearity/assay reportable range:

IgM Kappa: A linearity study was performed following CLSI *Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline (EP6-A)*. The linearity of this assay has been confirmed using a serially diluted serum sample over the range of 0.139–5.797 g/L with deviation from linearity < 10% using the 1 + 9 sample dilution.

IgM Lambda: A linearity study was performed following CLSI *Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved*

Guideline (EP06-A). The linearity of this assay has been confirmed using a serially diluted serum sample over the range of 0.164–4.979 g/L with deviation from linearity < 10% using the 1 + 9 sample dilution.

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 Optilite Hevylite IgM Kappa and Optilite Hevylite IgM Lambda kits:

A real-time stability study of unopened kits was performed on three lots of Optilite Hevylite IgM Kappa and IgM Lambda kits with testing time intervals at Day 0 and months 3, 6 and 10. Data support a shelf life claim of 9 months at 2–8° C. Real time stability study is on-going.

Open-vial stability was performed on three lots of Optilite Hevylite IgM Kappa and IgM Lambda kits with testing time intervals at Day 0 and months 1, 2, and 3. Data support the open vial stability claim of 3 months at 2–8° C.

On-board stability was performed on three lots of Optilite IgM Kappa kits with testing time intervals at 0, 14, 28, and 35 days and three lots of Optilite IgM Lambda kits with testing time intervals at 0, 22, 28, and 35 days. Data support the on-board stability claim of 30 days at 8–12° C, provided that the power is left switched on as stated in the product insert.

d. *Detection limit:*

Based on CLSI EP17-A *Protocols for Determination of Limits of Detection and Limits of Quantitation*.

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 was determined to be 0.0 g/L for both the Optilite Hevylite IgM Kappa kit and the Optilite Hevylite 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 0.0009 g/L for the Optilite Hevylite IgM Kappa kit and 0.0011 g/L for the Optilite Hevylite 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: for the

IgM κ kit LoD < LoQ and TE < 0.163 g/L; for the IgM λ kit LoD < LoQ and TE < 0.101 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 Optilite Hevylite IgM Kappa kit on the Optilite analyzer or the Optilite Hevylite IgM Lambda kit on the Optilite Analyzer. LoQ was calculated according to the formula TE = | Bias | + 2 x SDs. The LoQ was determined to be 0.02 g/L for the Optilite Hevylite IgM Kappa kit and 0.018 g/L for the Optilite Hevylite IgM Lambda kit.

Claimed Analytical sensitivity for each kit is summarized below:

Assay	LoB	LoD	LoQ
Optilite Hevylite IgM Kappa kit	0.0 g/L	0.0009 g/L	0.020 g/L
Optilite Hevylite IgM Lambda kit	0.0 g/L	0.0011 g/L	0.018 g/L

e. *Analytical specificity:*

Interfering Substances

A study was performed following CLSI EP7-A2: Interference Testing in Clinical Chemistry, Approved Guideline (CLSI Document EP7-A2). A normal serum sample, serum samples close to the medical decision points and abnormal serum samples were tested. No significant assay interference effects were observed when tested with intralipid (1500mg/dL), triglyceride (1000mg/dL), bilirubin (200mg/dL), haemoglobin (5g/L) or a range of commonly used therapeutic drugs as listed in the tables below:

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

Drug	Concentration tested
Acetaminophen	1324 μ mol/L
Acetylsalicylic Acid	3.63 mmol/L
Ascorbic Acid	342 μ mol/L
Bortezomib	6 mg/mL
Caffeine	308 μ mol/L
Cimetidine	79.2 μ mol/L
Cyclophosphamide	60 μ g/mL
Digoxin	7.8 nmol/L
Furosemide	90 μ mol/L

Ibuprofen	2425 µmol/L
Phenytoin	198 µmol/L
Methotrexate	2 mmol/L
Pomalidomide	100 µg/mL
Prednisolone	100 µg/mL
Theophylline	222 µmol/L

Although no interference was identified the package insert states that “turbidimetric assays are not suitable for measurement of highly lipemic or hemolyzed samples, or samples containing high levels of circulating immune complexes (CICs) due to the unpredictable degree of non-specific scatter these sample types might generate. Unexpected results should be confirmed using alternative assay method”.

Cross-reactivity:

Cross-reactivity studies were carried out by testing Optilite Hevylite IgM Kappa and Optilite 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 10 IgGκ, 6 IgGλ, 9 IgAκ, 8 IgAλ for the Optilite Hevylite IgM Kappa and 13 IgGκ, 9 IgGλ, 1 Biclinal IgGκ/IgMκ, 9 IgAκ, 6 IgAλ, multiple myeloma patient sera samples for the Optilite Hevylite IgM Lambda assays.

In addition, 12 IgMκ patient samples were tested on Optilite Hevylite IgM Lambda kits to investigate potential cross-reactivity, and similarly, 9 IgMλ patient samples were tested on IgM kappa kits.

No significant cross-reactivity was observed.

Antigen excess (hook effect):

The possibility of antigen excess occurring when using the device on The Binding Site Optilite analyzer was evaluated with 15 monoclonal IgM Kappa and 11 monoclonal IgM Lambda samples with concentrations above the 0.2–5.0 g/L and 0.18–4.5 g/L standard measuring ranges respectively. No antigen excess effect up to 48.0 g/L of IgMκ and 38.0 g/L of IgMλ concentration levels were observed at 1 + 9 sample dilution.

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:

IgM Kappa

A total of 263 samples spanning the measuring range of 0.024–51.75g/L were assayed in singlicate by both the Optilite Hevylite IgM Kappa kit and the Optilite Hevylite IgM Kappa Kit for use on SPA_{PLUS}. The serum samples included 24 normal donors, 23 Waldenström’s Macroglobulinemia patients and 216 (97 of which had elevated IgM Kappa) clinical samples that include IgM Kappa monoclonal gammopathy, IgM Lambda monoclonal gammopathy samples.

IgM Lambda

A total of 249 samples spanning the measuring range of 0.027–33.03g/L were assayed in singlicate by both the Optilite Hevylite IgM Lambda Kit and the Optilite Hevylite IgM Lambda Kit for use on SPA_{PLUS}. The serum samples included 24 normal donors and 22 Waldenström’s macroglobulinemia patients and 203 (39 of which had elevated IgM Lambda) clinical samples that include IgM Kappa monoclonal gammopathy, IgM Lambda monoclonal gammopathy samples.

IgM Kappa/ IgM Lambda Ratio

A total of 236 samples that gave both IgM Kappa and IgM Lambda results in the previous comparison studies were used to calculate the IgM Kappa/ IgM Lambda ratio for both the Optilite and the SPA_{PLUS} Hevylite IgM assays.

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	263	0.024–51.75	$y=0.93x-0.01$	0.91 to 0.95	-0.21 to 0.14
IgM Lambda	249	0.027–33.03	$y=1.01x + 0.00$	0.98 to 1.03	-0.01 to 0.00
IgM κ /IgM λ ratio	236	0.006–858.95	$y= 0.87x + 0.13$	0.86 to 0.90	0.09 to 0.18

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):

Not applicable

4. Clinical cut-off:

Not applicable.

5. Expected values/Reference range:

The reference interval ranges below were obtained by measuring the IgM kappa and IgM lambda concentration of serum taken from 147 healthy adult UK blood donors using this assay on SPA_{PLUS}® (K140686). The reference interval was calculated using non-parametric statistics and represents the central 95% of the population. This was verified on the Optilite Hevylite using sera from 50 normal US blood donors. The study was based on CLSI C28-A3: *Defining, Establishing and Verifying Reference Intervals*.

Reference interval range results as included in the package insert:

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
IgMκ/ IgMλ ratio	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.