510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:

K183151

B. Purpose for Submission:

New Device

C. Measurand:

IgA

D. Type of Test:

Quantitative, Immunoturbidimetry

E. Applicant:

The Binding Site Group, Ltd.

F. Proprietary and Established Names:

Optilite IgA CSF 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:

   CFN – Method, Nephelometric, Immunoglobulins (G, A, M)

4. Panel:

   Immunology (82)

H. Intended Use:

1. Intended use:

   The Optilite IgA CSF Kit is intended for the quantitative *in vitro* measurement of IgA in cerebrospinal fluid (CSF) using the Optilite analyser.

2. Indication for use:

   Same as intended use.

3. Special conditions for use statement:

   For prescription use only.

4. Special instrument requirements:

   The Binding Site Optilite analyser

I. Device Description:

The Optilite IgA CSF Kit comprises the following reagents:

Latex Reagent: Supplied in stabilised liquid form. Preservatives: 0.025% sodium azide, 0.1% E-amino-n-caproic acid (EACA) and 0.01% benzamidine, 0.05% ProClin.

Calibrator and Controls: Pooled human serum, supplied in stabilised liquid form containing 0.099% sodium azide, 0.1% EACA and 0.01% benzamidine as preservatives. The concentration given on the quality control certificate has been obtained by comparison with the DA470k international reference material.

Reaction Buffer: Contains 0.099% sodium azide as a preservative.

J. Substantial Equivalence Information:

1. Predicate device name:

   Beckman Coulter IMMAGE Immunochemistry System Low Concentration Immunoglobulin A (IGALC) Reagent and Cerebrospinal Fluid Protein Calibrator (CSF CAL)

2. Predicate 510(k) number:

   K993549

3. Comparison with predicate:

Similarities		
Item	Device Optilite IgA CSF Kit	Predicate Beckman Coulter IMMAGE Immunochemistry System Low Concentration Immunoglobulin A (IGALC) Reagent and Cerebrospinal Fluid Protein Calibrator (CSF CAL)
Intended Use	The Optilite IgA CSF Kit is intended for the quantitative *in vitro* measurement of IgA in cerebrospinal fluid (CSF) using the Optilite analyser.	The IMMAGE Immunochemistry System Low Concentration Immunoglobulin A (IGMAC) Reagent, when used in conjunction with Beckman Coulter's IMMAGE Immunochemistry Systems and Cerebrospinal Fluid Protein Calibrator, is intended for the quantitative determination of human immunoglobulin A in serum and cereprospinal fluid b rate nephelometry.
Analyte	IgA	Same
Traceability	ERM-DA470k/IFCC	Same
Reagent type	Latex enhanced	Same
Reference interval	< 2.0 mg/L	Same
Assay type	Quantitative	Same

Differences		
Item	Device Optilite IgA CSF Kit	Predicate Beckman Coulter IMMAGE Immunochemistry System Low Concentration Immunoglobulin A (IGALC) Reagent and Cerebrospinal Fluid Protein Calibrator (CSF CAL)
Calibrator	Stabilized human serum	Human urine
Open vial stability	Three months	30 days
Antibody	Sheep anti-human IgA, polyclonal	Goat anti-human IgA
Method	Turbidimetric	Nephelometric
Measuring range	0.91 – 20 mg/L (1+0) 1.65 – 40 mg/L (1+1)	Initial: 2 – 70 mg/L Extended: 0.25 – 420 mg/L
Instrument	Binding Site Optilite	Beckman Coulter IMMAGE
Specimen type	CSF	CSF (and serum)

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

CLSI EP17-A2 Protocols for Determination of Limits of Detection and Limits of Quantitation; Approved Guideline

CLSI EP07-A2 Interference Testing in Clinical Chemistry, Approved Guideline – Second Edition

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

CLSI EP05-A3 Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline – Third 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:

1. Analytical performance:

The results of all the studies met the Manufacturer's pre-specified acceptance criteria.

a. Precision/Reproducibility:

The precision study was performed over five working days, with two runs per day. The study was conducted with two users, using two reagent lots, each carried out on three analyzers. Level 1 sample was contrived by adding native CSF to a mock CSF matrix. Level 4 sample was contrived by adding purified IgA protein to native CSF. The purified IgA was manufactured in house from a pool of normal human serum. The IgA protein was purified by anti-IgA affinity chromatography, eluted with a buffer and then run through an adsorption column to complete the process. Level 2 and 3 samples were native CSF. Kit lots 1 and 2 were used for Levels 2, 3 and 4. Kit lots 3 and 4 were used for Level 1.

Results are summarized below:

Within-Laboratory Imprecision:

Level	N	Mean (mg/L)	Within-Run		Between-Run		Between-Day		Total	
			SD	%CV	SD	%CV	SD	%CV	SD	%CV
1	**20**	2.235	0.028	1.3	0.042	1.9	0.073	3.3	0.089	4.0
2	**20**	3.526	0.087	2.5	0.082	2.3	0.157	4.4	0.197	5.6
3	**20**	4.586	0.035	0.8	0.049	1.1	0.195	4.2	0.204	4.4
4	**20**	28.893	0.479	1.7	0.556	1.9	0.403	1.4	0.837	2.9

Instrument-to-Instrument Imprecision:

Level	Mean (mg/L)	Between lot		Between instrument	
		SD	%CV	SD	%CV
1	2.235	0.011	0.5	0.057	2.5
2	3.526	0.040	1.1	0.172	4.9
3	4.586	0.076	1.7	0.203	4.5
4	28.893	0.110	0.4	0.622	2.2

b. *Linearity/assay reportable range:*

The linearity study was conducted using mock CSF samples that covered the range of the standard sample dilution (1+1) from 1.65 mg/L to 40 mg/L. Mock CSF samples were contrived by spiking in purified IgA into a mock CSF matrix. The analyte concentrations of the high and low mock CSF samples were selected in order to cover at least 10% beyond the limits of the measuring range of the assay (1.025 – 47.876 mg/L). A dilution series (n=11) was prepared by blending the high pool and low pool. Three replicates of each level of the dilution series were run and the mean value calculated. Data were analyzed using linear regression and deviation from linearity according to CLSI guideline EP6-A. The results demonstrate that the assay is linear over the range of 1.025 – 47.876 mg/L at the standard 1+1 sample dilution. This means the analytical measuring interval is linear over the range 0.83 – 20 mg/L (neat, 1+0), and confirms that the assay is linear over the width of the curve at all available sample dilutions.

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

Traceability:

The calibration of the assay is traceable to ERM-DA470k/IFCC.

Stability:

Real-time stability: The purpose of this study was to validate the stability of the Optilite IgA CSF kit when stored at the recommended storage temperature of 2–8°C for the duration of the kit shelf life. The study was performed with one master calibrator (1923 mg/L), low control (4.614 mg/L) and high control (27.068 mg/L) using three reagent lots at time points (months) of 0, 3, 6.5, 10, 13 and 19. The Optilite IgA CSF Reagent, Calibrator and Controls have a shelf life of up to 18 months.

Open-vial stability: The purpose of this study was to validate the open-vial stability of the Optilite IgA CSF kit reagents when stored at 2–8°C for three months as stated in the product insert. Two samples were used for this study, a low control (4.61 mg/L), and high control (27.068 mg/L), which were run on days 0, 32, and 111 for lot 1, on days 0, 28, 56 and 107 for lot 2, and on days 0, 32, 60 and 111 for lot 3. The Optilite IgA CSF Reagent, Calibrator and Controls can be stored opened at 2–8°C for up to 3

months.

On-board stability: The purpose of this study was to confirm the stability of the Optilite IgA CSF kit, once opened and stored 'on-board' the Optilite analyser for a period of at least 30 days. Two samples were used for this study, a low control (4.61 mg/L), and high control (27.068 mg/L), which were run on days 0, 8, 15, 22, 29 and 36 for lot 1, on days 0, 7, 14, 21, 28 and 35 for lot 2, and on days 0, 7, 13, 21, 28 and 35 for lot 3. The Optilite IgA CSF Reagent can be stored on-board the Optilite Analyser for up to 30 days.

d. Detection limit:

The purpose of this analytical sensitivity study was to estimate the Limit of Blank (LoB) and Limit of Detection (LoD) and to validate the Limit of Quantitation (LoQ) of the Optilite IgA CSF Kit, to ensure that these meet the pre-defined criteria. The study was carried out using mock CSF samples designed to mimic patient samples as closely as possible.

LoB: The LoB study was carried out using four samples of mock CSF matrix and two reagent lots. Samples were run five times each per day, over 3 days to give a total of 60 replicates per reagent lot. For each reagent lot, 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. The LoB for the assay was determined to be 0.283 mg/L.

LoD: Four mock CSF sample matrices spiked with purified IgA were used to calculate the LoD using two reagent lots. Samples were run five times each per day, over 3 days. Samples were targeted so that they were close to the bottom of the measuring range (0.91 mg/L) for the neat (1+0) dilution. The LoD was calculated for each reagent lot. The LoD for the assay was determined to be 0.3562 mg/L.

LoQ: Five mock CSF sample matrices spiked with purified IgA with concentrations close to the bottom of the standard measuring range and two reagent lots were used for this study. These samples were run five times over 5 days. The LoQ was validated and set to be the bottom of the measuring range, 0.9100 mg/L.

e. Analytical specificity:

Interference:

Two samples were tested: mock CSF matrix spiked with purified IgA (2.184 mg/L, and 5.239 mg/L), with clinically relevant concentrations to evaluate the effects of hemoglobin, bilirubin, acetaminophen and acetylsalicylic acid on the results of the assay. The two contrived samples were spiked separately with the interferents and with appropriate blank material to create 'negative' control samples. No significant assay interference effects were observed in CSF when tested with hemoglobin

(2.5g/L), bilirubin (100mg/L), acetaminophen (1324µmol/L) or acetylsalicylic acid (3.63mmol/L).

Antigen Excess:

Turbidimetric assays can be susceptible to antigen excess with high concentration samples resulting in a falsely low result. This study is designed to investigate the antigen excess capacity of the Optilite IgA CSF kit, demonstrating that the native antigen excess capacity is robust enough to ensure that very high concentration samples flag correctly as "high activity". This error flag means the sample is in antigen excess. The antigen excess capacity is defined as the concentration at which the resultant change in Optical Density (OD) is not reported lower than the OD given for the standard top calibrator. Antigen excess capacity was determined at neat dilution. A calibration curve was extended with the addition of three extra calibrator points; at 53.14 mg/L, 59.05 mg/L and 65.61 mg/L. The additional calibrator points were manufactured using IgA calibrator material diluted in analyzer diluent. This curve spanned a measuring range of 1.69 – 65.61 mg/L. The absorbance value for each calibrator point was analyzed to determine the antigen excess capacity of the assay. The concentration at which the absorbance value is not reported as lower than the value given by the standard calibrator is the demonstrated antigen excess capacity of the assay. This study showed that the Optilite IgA CSF kit has sufficiently robust antigen excess capacity up to at least 65.61 mg/L at the 1+1 sample dilution, which is equivalent to 1.64 times the top point of the standard calibration curve.

f. Assay cut-off:

Refer to expected values

2. Comparison studies:

a. Method comparison with predicate device:

A comparison study was performed by analyzing 130 CSF samples using the Optilite IgA CSF Kit and in comparison to the predicate. A total of 120 native samples were tested in this study in order to cover the measuring range, and these samples were from patients with various admission diagnoses. Additionally, 10 patient samples were spiked with purified IgA in order to cover the upper end of the measuring range as it was not possible to source native CSF samples with sufficiently high concentrations of IgA. Samples were stored at −20°C before testing and in accordance with the recommendations in the product insert. Eight samples were excluded from the analysis as they were below the test assay measuring range.

Passing Bablok regression analysis generated the following results:

Analyte	N	Sample Range	Passing & Bablok	Slope 95% CI	Intercept 95% CI
IgA	122	0.96–39.44 g/L	y = 1.05x – 0.02	1.03 to 1.07	-0.07 to 0.05

The measured correlation coefficient was 0.984.

All samples were analyzed in singlicate on the new device and the predicate device. Overall, the results of the method comparison study demonstrate that the test device correlates well with the predicate in terms of overall concordance. Concordance analysis demonstrated 96.7% positive agreement (95% CI = 90.6 – 99.3), and 97.5% negative agreement (95% CI = 86.8 – 99.9%) between samples. Overall, 96.9% of samples gave concordant results between test and predicate assays.

For additional information on the clinical samples used in the method comparison study, see "Other clinical supportive data" section below.

b. Matrix comparison:

Not applicable

3. Clinical studies:

a. Clinical Sensitivity:

Not applicable

b. Clinical specificity:

Not applicable

c. Other clinical supportive data (when a. and b. are not applicable):

Individual sample demographic information and admission diagnosis for the samples used in the method comparison study, where available, are summarized below:

N	Age (available for 39/120 samples)		Gender (available for 105/120 samples)		
	Range	Median	Male	Female	Unknown
120	22–87	44	45	60	15

Admission diagnosis information:

Group	Admission Diagnosis	Number
Neurological	ADEM	1
	Neuropathy	1
	Neurodegeneration	1
	Idiopathic Intracranial Haemorrhage 1	1
	Autoimmune encephalitis	2
	Optic neuritis	1
	Neuroinflammation	1
	Myelitis	1
	CNS disorder	6
	Multiple Sclerosis	1
Other		7
Unknown		97

4. Clinical cut-off**:**

   Refer to expected values

5. Expected values/Reference range:

   The reference range of <2.0 mg/L was transferred from literature as was done for the predicate device.

N. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Parts 801 and 809, as applicable.

O. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.