

**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION MEMORANDUM
ASSAY ONLY TEMPLATE**

A. 510(k) Number:

k142735

B. Purpose for Submission:

New Device

C. Measurand:

IgG3 subclass Antibody

D. Type of Test:

Quantitative, turbidimetric

E. Applicant:

The Binding Site Group, Ltd.

F. Proprietary and Established Names:

Optilite® IgG3 Kit

G. Regulatory Information:

1. Regulation section:

21 CFR §866.5510 – Immunoglobulins A, G, M, D, 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(s):

The Optilite IgG3 kit is intended for the quantitative *in vitro* measurement of human IgG3 in serum, using the Binding Site Optilite analyser. Measurement of this immunoglobulin is an aid in the diagnosis of abnormal protein metabolism and the body's lack of ability to resist infectious agents. The test result should be used in conjunction with other laboratory and clinical findings.

2. Indication(s) for use:

Same as Intended Use

3. Special conditions for use statement(s):

For prescription use only

4. Special instrument requirements:

Optilite analyzer (k141100)

I. Device Description:

The device consists of the following: polyclonal monospecific sheep anti-IgG3 antisera coated onto polystyrene latex in liquid form in the presence of preservatives (0.05% ProClin™; 0.033% sodium azide; 0.1% E-amino-n-caproic acid (EACA); 0.01% benzamidine); IgG3 calibrator and Controls (Low, High and Elevated levels) in stabilized liquid form (with preservatives: 0.099% sodium azide, 0.1% EACA and 0.01% benzamidine); Reaction Buffer (with 0.099% sodium azide as preservative); and Optilite Diluent 1.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Binding Site Human IgG and IgG subclass (IgG1, IgG2, IgG3, IgG4) liquid reagent kits for use on the SPAplus

2. Predicate 510(k) number(s):

k072889

3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Assay type	Quantitative	Same
Detection Method	Turbidimetric immunoassay	Same
Sample matrix	Serum	Same
Antibody	Polyclonal monospecific sheep anti-human IgG3 (F(ab) ₂) fragment bound to 200 nm latex particles	Same
Traceability	Standardized against ERM-DA470k European Reference Material (previously CRM470)	Same
Open vial stability	3 months	Same
On board vial stability	30 days	Same
Reference Ranges (mg/dL) (95% percentile range)	Adults: 218.2-1760.6 Pediatric: 0-2 yrs: 186-853 2-4 yrs: 173-676 4-6 yrs: 99-1221 6-8 yrs: 155-853 8-10 yrs: 127-853 10-12 yrs: 173-1730 12-14 yrs: 283-1250 14-18 yrs: 230-1096	Same

Differences		
Item	Device	Predicate
Intended Use	Quantitative measurement of IgG3 in serum	Quantitative measurement of Human IgG and IgG subclasses IgG1, IgG2, IgG3, IgG4 in serum
Calibrator	One Optilite IgG3 Calibrator	Six Calibrators for each: IgG, IgG1, IgG2, IgG3, IgG4
Controls	Low, High and Elevated levels, liquid, ready-to-use	Low and high IgG and IgG subclasses: IgG1, IgG2, IgG3, IgG4
Instruments	Optilite® Analyzer	SPA _{PLUS} TM Analyser
Measuring range (mg/dL)	5.5-220 (1:2 dilution) 55-2200 (1:20 dilution)	55-1000 (1:10 dilution) 220-4000 (1:40 dilution)

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

- CLSI EP05-A2: Evaluation of Precision Performance of Quantitative Measurement Methods; Approved Guideline – Second Edition
- CLSI EP06-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach
- CLSI EP07-A2: Interference Testing in Clinical Chemistry
- CLSI EP17-A: Protocols for Determination of Limits of Detection and Limits of Quantitation
- CLSI C28-A3: Defining, Establishing and Verifying Reference Intervals in the Clinical Laboratory

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, a portion of the light is transmitted and focused onto a photo iodide 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 within-run, between-run, between-day, between-lot and between-instrument precision were determined by testing five serum samples over 21 days with two runs per day on three different reagent lots on three analysers. Results are summarized below.

Precision Summary													
	Mean (mg/L)	Within run		Between run		Between day		Between lot		Between instrument		Total	
		SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV	SD	%CV
Level 1*	9.1	0.8	8.4	0.0	0.0	0.8	8.8	0.1	1.6	0.5	5.5	1.1	12.1
Level 2	118.5	6.0	5.1	0.0	0.0	7.8	6.6	3.1	2.6	2.6	2.2	9.9	8.3
Level 3	164.3	5.0	3.0	1.5	0.9	11.2	6.8	5.8	3.3	0.6	0.1	12.3	7.5
Level 4	256.5	9.7	3.8	3.0	1.2	13.7	5.3	7.6	3.0	4.0	1.6	17.0	6.6
Level 5	1785.4	35.3	2.0	34.4	1.9	91.1	5.1	38.9	2.2	55.3	3.1	103.6	5.8

*performed at the 1:2 dilution

b. Linearity/assay reportable range:

A linearity study was performed following CLSI document Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach, Approved Guideline (EP6-A). The linearity of this assay has been confirmed using serially diluted serum samples to cover the range 5.4 – 216.4 mg/L for Optilite Analyzer Dilution 1+1 and 54.1 – 2164.4 mg/L for Optilite Analyzer Dilution 1+19 with deviation from linearity $\leq 9.9\%$. Regression equation for the high linear range (280.27-2477.50 mg/L: $y=0.99x + 4.36$; $r = 1.000$ and for the low linear range (38.38-677.14 mg/L: $y=1.01x + 1.52$; $r = 1.000$).

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

An Internal Reference standard (IR) was assigned by comparison with the European Reference Material ERM-DA470k. Value assignment from total IgG to IgG3 was performed following the protocol outlined in Carr-Smith, H.D. et al. IgG subclass value assignment to protein reference preparation CRM470. Clin. Chem. Vol. 43, No.56, PS238 and Williams, D.R. et al. Assignment of IgG Subclass values to the Protein Reference Preparation DA470k. Clin. Chem. Vol. 55, No. S6, PS C-90.

Stability:

A real-time stability study was performed on three lots of Optilite IgG3 kit with testing time intervals at day 0, 3 months and 6.5 months. Data support a shelf life claim of 6 months at 2-8°C.

Open kit stability was performed on three lots of OptiliteIg3 kit with testing time intervals at day 0, 1 week and 3 months. Data support the open kit stability claim of 3 months at 2-8°C.

Throughout this study the kit components were stored at the recommended storage temperature of 2-8°C.

On-board stability was performed on three lots of OptiliteIg3 kit with testing time intervals at day 0, 14, 21, 28, and 35. Data support the on-board stability claim of 30 days at 8-12°C.

All stability results were within the sponsor's acceptance criteria. Real time stability study is on-going.

d. Detection limit:

The analytical sensitivity was determined in accordance with CLSI EP17-A2. The Limit of Blank (LoB) was based on 60 determinations of a blank sample and was estimated at 0.92 mg/L. The Limit of Detection (LoD) was calculated at 1.40 mg/L according to the equation: the LoB + 1.645 x SDs where SDs, the standard deviation, was based on 60 determinations of a sample with analyte level near the lower limit of the reportable range. The LoQ was calculated at 5.50 mg/L and the

bias was 0.86 mg/L. The tabulated summary is shown below:

LoB	LoD	LoQ
0.92mg/L	1.40mg/L	5.50mg/L

e. *Analytical specificity:*

Interference by endogenous and other substances:

No significant assay interference ($\pm 10\%$) by triglycerides (1000 mg/dL), Intralipid (2000 mg/dL), 200 mg/L bilirubin, or 5g/L hemoglobin using IgG3 samples at 115.7 mg/L, 245.1 mg/L and 1188.9 mg/L.

No significant assay interference by the following 16 therapeutic drugs:

Drug	Concentration tested
Acetaminophen	1324 μ mol/L
Acetylsalicylic Acid	3.63mmol/L
Amoxicillin	206 μ mol/L
Ascorbic Acid	342 μ mol/L
Caffeine	308 μ mol/L
Cefotaxime	673 μ mol/L
Cefuroxime	1416 μ mol/L
Chloramphenicol	155 μ mol/L
Cimetidine	79.2 μ mol/L
Digoxin	7.8nmol/L
Fluconazole	245 μ mol/L
Ibuprofen	2425 μ mol/L
Penicillin	75mg/L
Phenytoin	198 μ mol/L
Theophylline	222 μ mol/L
Vancomycin	69 μ mol/L

The package insert states that “turbidimetric assays are not suitable for measurement of samples containing rheumatoid factor or paraproteins, highly lipemic or hemolyzed samples, or samples containing high levels of circulating immune complexes due to the unpredictable degree of non-specific scatter these sample types might generate. Unexpected results should be confirmed using alternative assay method”.

Antigen excess effect:

The possibility of antigen excess occurring when using the device on The Binding Site Optilite® was evaluated with samples with IgG3 concentrations above the assay range (2500, 2900, and 3300 mg/L). No antigen excess effect up to 3300 mg/L of IgG3 was observed.

The package insert states that “potential occurrences of antigen excess cannot be

completely excluded; in rare cases samples with monoclonal IgG3 may give falsely low results due to antigen excess. Where this is possible or suspected it is recommended that the samples are re-assayed at a higher dilution to confirm result.”

f. Assay cut-off:

Not applicable.

2. Comparison studies:

a. Method comparison with predicate device:

A method comparison study between Optilite IgG3 Kit and predicate device was performed by analyzing 293 samples including 26 normal and 267 clinical sera (22 hypergammaglobulinemia, 2 hypogammaglobulinemia, 13 suspected hypergammaglobulinemia, 6 suspected hypogammaglobulinemia, and 224 unknown diagnosis).

Passing-Bablok regression analysis generated the following result: $y = 1.04x - 6.04$ mg/L with correlation coefficient $r = 0.991$.

The package insert states that “diagnosis cannot be made and treatment must not be given on the basis of IgG3 measurements alone. Clinical history and other laboratory findings must be taken into account”.

b. Matrix comparison:

Not applicable

3. Clinical studies:

a. Clinical Sensitivity and specificity:

Not applicable

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

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference Interval:

Adult reference interval: 218.2-1760.6 mg/L

Pediatric reference interval is as follows:

Age Group (years)	Number (n)	95% Reference Interval (mg/L)
0 – 2	39	186 - 853
2 – 4	36	173 – 676
4 – 6	49	99 – 1221
6 – 8	43	155 – 853
8 – 10	32	127 – 853
10 – 12	46	173 – 1730
12 – 14	54	283 – 1250
14 – 18	48	230 - 1096

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.