

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

A. 510(k) Number:

k083289

B. Purpose for Submission:

New device

C. Measurand:

Beta-2 Microglobulin

D. Type of Test:

Quantitative, latex-enhanced turbidimetric assay

E. Applicant:

The Binding Site, Ltd.

F. Proprietary and Established Names:

Human Beta-2 Microglobulin Kit for use on SPA_{PLUS}TM

G. Regulatory Information:

1. Regulation section:

21 CFR § 866.5630 Beta-2 microglobulin immunological test system

2. Classification:

Class II

3. Product codes:

JZG, Beta-2 Microglobulin, Immunological Test System

4. Panel:

Immunology 82

H. Intended Use:

1. Intended use(s):

The kit is intended for the quantitative *in vitro* determination of beta-2 microglobulin (β2M) in human serum and urine, using SPA_{PLUS}TM analyser to aid the diagnosis of active rheumatoid arthritis and kidney disease. The test result is to be used in conjunction with other clinical and laboratory 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:

SPA_{plus}TM analyser

I. Device Description:

The device consists of the following: monospecific sheep anti-β2M antibody coated onto polystyrene latex in the presence of preservatives. Calibrators 1-6; low, high and elevated controls in liquid form; and β2M reaction buffer. The reagents contain 0.05% ProclinTM, 0.099% sodium azide, 0.1% E-amino-n-caproic acid (EACA) and 0.01% benzamidine as preservative.

J. Substantial Equivalence Information:

1. Predicate device name(s):

The Binding Site β2M on the BNII

2. Predicate K number(s):

k946069

3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	Quantitative determination of β 2M in serum or urine to aid in the diagnosis of active rheumatoid arthritis and kidney disease	Same
Detection Method	Turbidimetric immunoassay	Same
Traceability	Standardized against the 1 st International Standard for Beta-2 Microglobulin (NIBSC)	Same
Sample Matrix	Human serum and urine	Same
Antibodies	Monospecific Sheep	Same

Differences		
Item	Device	Predicate
Controls	Low, High and Elevated levels liquid ready to use	Low, High and Elevated levels lyophilized
Instruments	SPA _{PLUS} TM analyser	BNII analyser
Measuring range	Serum: 0.3 - 40 mg/L Urine: 0.03 - 20 mg/L	Serum: 0.7 – 22.5 mg/L Urine: 0.04 mg/L
Reference Range	Serum: 0.80 – 2.34 mg/L Urine: 0.030 – 0.202 mg/L	Serum: 1.22 – 2.46 mg/L Urine: <0.03– 0.23 mg/L

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

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

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.

Latex-enhanced antibodies. Some antigen-antibody reactions do not form sufficiently large immune complexes to be detected turbidimetrically. If the antibody is coated

onto latex particles of a suitable size, the light scattering ability of the immune complexes formed with antigen is enhanced sufficiently to enable turbidimetric detection.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

The intra-assay precision was determined by testing six serum samples twenty one times. The inter-assay precision was determined by testing three serum samples three times with two runs per day for 21 days. Results are summarized below.

Intra-assay:

n=21	β2M		
	SD	Concentration (mg/L)	% CV
Sample 1	0.02	0.97	1.8
Sample 2	0.03	4.94	0.6
Sample 3	0.09	16.43	0.6
Sample 4	0.035	20.74	1.7
Sample 5	0.065	31.81	2.0
Sample 5	0.62	36.53	1.7

Inter-assay:

n=21	β2M		
	SD	Concentration (mg/L)	% CV
Sample 1	0.06	0.97	5.8
Sample 2	0.10	4.94	2.1
Sample 3	0.31	16.43	1.9

b. *Linearity/assay reportable range:*

Serum linearity across the assay range (0.3- 40.0 mg/L) was confirmed by testing three sera with concentrations from 9.17- 36.46 mg/L. The samples were serially diluted 10 times with buffer (1:10) down to the lower measuring range (0.3 mg/L). All testing were performed twice. The regression plot equations where y is the measured level of serum β2M concentration and x the theoretical concentration were:

$$y = 0.9896x + 0.150 \text{ (mg/L)}, r^2 = 0.9996 \text{ for serum } \beta 2M$$

Urine linearity across the assay range (0.6-20.0 mg/L) was confirmed by testing two samples with concentrations of 0.853 mg/L and 18.300 mg/L. The samples were serially diluted off-line 10 times with buffer (1:20) down to the lower measuring range (0.04 mg/L). All testing were performed three times. The regression plot equations where y is the measured level of urine β2M concentration and x the theoretical concentration were:

$$y = 0.9929x - 0.069 \text{ (mg/L)}, r^2 = 0.9992 \text{ for urine } \beta 2\text{M}$$

- c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*
An internal reference standard (IR8494) was assigned by comparison to the 1st International Standard for Beta-2 Microglobulin (NIBSC Code: B2M).

Stability: The expiration date claims are 6 months for the $\beta 2\text{M}$ unopened Kit and 30 days for the opened $\beta 2\text{M}$ kit and on-board $\beta 2\text{M}$ kit.

- d. *Detection limit:*

The detection limit was determined by testing a blank sample, the lowest calibrator, and a sample with value close to the blank sample (0.006 mg/L) 60 times each. The very low level sample (diluted low level urine) gave a mean value of 0.00672 absorbance unit giving an estimated value of 0.12 mg/L which is considered to be the limit of detection. The limit of quantitation for this assay is defined as the lowest point of the calibration curve: 0.03 mg/L based upon a neat sample dilution (minimum used for urine).

- e. *Analytical specificity:*

Serum Interference by endogenous and other substances: A known $\beta 2\text{M}$ serum sample was tested with the following interferents: 5 g/L hemoglobin, 200 mg/L bilirubin, 10722 FTU of chyle and 600 IU/mL RF. No significant interference by these substances was observed.

Urine Interference by endogenous and other substances: A known $\beta 2\text{M}$ urine sample was tested in with the following interferents: 238 m g/L hemoglobin, 200 mg/L bilirubin, 200 mg/L ascorbic acid, and 100 mg/dL therapeutic IgG. No significant interference by these substances was observed.

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 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 SPA_{PLUS}™ was evaluated with samples with $\beta 2\text{M}$ concentration above the assay range (40 mg/L). No antigen excess effect up to 76 mg/L of $\beta 2\text{M}$ was observed. The package insert states: “The antigen excess protection (P flag) on the SPA_{PLUS} has been tested up to a level which is equivalent to 760 mg/L.

- f. *Assay cut-off:*

Not provided

2. Comparison studies:

- a. *Method comparison with predicate device:*

Testing was performed on 103 sera (31 normal and 72 clinical) and 49 urine

samples (24 normal and 25 known renal impaired samples).

The table below shows the comparison of 103 sera (ranging from 0.51 – 19.44 mg/L) and 49 urine samples (ranging from 0.04- 20.2 mg/L) that were tested with the Binding Site SPA_{PLUS}TM β2M assay and the predicate device Behring BNII System. Regression analysis of these samples is summarized below:

The Binding Site SPA _{PLUS} TM vs. Behring BNII analyzer	n	Regression Equation	r
Serum	103	$y = 1.02x - 0.11$ (mg/L)	0.996
Urine:	49	$y = 0.97x + 0.00$ (mg/L)	0.993

b. Matrix comparison:

Not applicable.

3. Clinical studies:

a. Clinical Sensitivity and specificity:

None provided.

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

Not applicable.

4. Clinical cut-off:

Not provided

5. Expected values/Reference range:

Adult normal serum range was assessed on a total 150 normal adult sera samples from healthy US blood donors (age 18-64 years old). The urine range was assessed on a total of 116 normal urines from UK healthy adult donors (age 18-65 years old). The assays were performed on the Binding Site SPA_{plus}TM analyser. A non-parametric distribution of β2M results were seen that gave a 95 percentile reference interval of 0.8-2.34 mg/L with a mean of 1.33 mg/L and a median of 1.26 mg/L for serum; and 95 percentile reference interval of 0.03-0.202 mg/L with a mean of 0.08 mg/L and a median of 0.07 mg/L for urine.

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.