

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

A. 510(k) Number:

k143118

B. Purpose for Submission:

New device

C. Measurand:

Urine Albumin

D. Type of Test:

Quantitative turbidimetric immunoassay

E. Applicant:

The Binding Site Group, Ltd.

F. Proprietary and Established Names:

Human Microalbumin Kit for use on SPA_{plus} Kit

G. Regulatory Information:

| Product Code | Classification | Regulation | Panel |
|--------------|----------------|---|--------------------|
| DCF | Class II | 21 CFR §866.5040 Albumin Immunological Test System | Immunology (82) |

H. Intended Use:

1. Intended use(s):

See indications for use below.

2. Indication(s) for use:

The Human Microalbumin Kit for use on SPA_{PLUS} kit is intended for the quantitative measurement of human albumin in human urine using the SPA_{PLUS} turbidimetric analyzer. Measurement of albumin in human urine is an aid in the diagnosis of renal disease in conjunction with other laboratory and clinical findings. For in vitro diagnostic use only.

3. Special conditions for use statement(s):

Prescription use only.

4. Special instrument requirements:

SPA_{PLUS} turbidimetric analyzer

I. Device Description:

The Human Microalbumin Kit for use on SPA_{plus} contains the following components:

- Human Microalbumin Antiserum SPA_{plus}: This antiserum is monospecific for albumin and is supplied in stabilized liquid form.
- Human Microalbumin SPA_{plus} Calibrator Set 1-6 and Human Microalbumin SPA_{plus} High & Low Controls. The calibrators and controls consist of pooled human serum and are supplied in stabilized liquid form. The calibrators are supplied as calibrators A through F targeting 6 concentrations of albumin. The controls are supplied as a low concentration and high concentration control. Calibrators and controls are provided ready to use with the Human Microalbumin Kit.
- Microalbumin Reaction Buffer SPA_{plus}: Contains 0.099% sodium azide, as a preservative.

Each human donor unit used to manufacture the calibrators and controls were tested using FDA-accepted methods and found nonreactive for Hepatitis B Surface Antigen (HbsAg), antibody to Hepatitis C (HCV) and antibody to HIV-1/HIV-2.

J. Substantial Equivalence Information:

1. Predicate device name(s):

Siemens N Antiserum to Human Albumin

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

k972929

3. Comparison with predicate:

| Similarities | | |
|---------------------|--|--|
| Item | Proposed device | Predicate device (k972929) |
| Intended Use | Intended for quantitative measurement of albumin in urine as an aid in diagnosis of renal disease. | Intended to be used in the measurement of albumin in urine as an aid in the diagnosis of kidney and intestinal diseases. |
| Analyte | Albumin | same |
| Measuring Range | 11-344 mg/L | same |

| Differences | | |
|--------------------|-------------------------------|---|
| Item | Proposed device | Predicate device (k972929) |
| Antibody | Sheep anti-human Microalbumin | Rabbit anti-human Microalbumin |
| Principle | Turbidimetry | Nephelometry |
| Specimen Type | Urine | CFS, serum, heparinized and EDTA plasma and urine |

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

CLSI EP05-A2: Evaluation of Precision Performance of Quantitative Measurement Methods

CLSI EP06-A: Evaluation of Linearity of Quantitative Measurement Procedures

CLSI EP07-A2: Interference Testing in Clinical Chemistry

CLSI EP17-A: Protocols for Determination of Limits of Detection and Limits of Quantitation

L. Test Principle:

The Binding Site Group Human Microalbumin Kit for use on SPA_{Plus} test system is an immunoturbidimetric assay. Albumin agglutinates with anti-albumin antibodies (sheep). The precipitate is determined turbidimetrically by the amount of transmitted light, which is indirectly proportional to the specific albumin 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:

All performance testing was performed using the SPA_{Plus} analyzer.

a. *Precision/Reproducibility:*

The precision of the Human Microalbumin Kit on the SPA_{plus} analyzer was evaluated using a 21 day study performed in-house based upon the CLSI EP5-A2 Guideline using a panel of human urine samples. The study was performed using the following 5 samples: normal, Level 1 (negative sample); 25% below the medical decision point, Level 2; 25% above the medical decision point, Level 3; moderate pathological sample, Level 4 and high pathological sample, Level 5. The study was performed with 3 reagent lots each and was performed on 5 SPA_{plus} Analyzers. Samples were tested in duplicate, twice a day, over 21 days (N = 84).

Microalbumin Precision Study Results:

| Level | Mean (mg/L) | Within Run | | Between Run | | Between Day | | Total | |
|-------|-------------|------------|-----|-------------|-----|-------------|-----|-------|-----|
| | | SD | %CV | SD | %CV | SD | %CV | SD | %CV |
| 1 | 20.14 | 0.33 | 1.7 | 0.47 | 2.3 | 1.08 | 5.4 | 1.22 | 6.1 |
| 2 | 23.79 | 0.36 | 1.5 | 1.70 | 7.1 | 1.29 | 5.4 | 2.16 | 9.1 |
| 3 | 37.54 | 0.17 | 0.5 | 1.00 | 2.7 | 1.44 | 3.8 | 1.76 | 4.7 |
| 4 | 144.18 | 2.69 | 1.9 | 1.38 | 1.0 | 9.74 | 6.8 | 10.19 | 7.1 |
| 5 | 281.57 | 6.05 | 2.1 | 4.55 | 1.6 | 21.83 | 7.8 | 23.10 | 8.2 |

| Level | Mean (mg/L) | Between Lots | | Between Instrument | |
|-------|-------------|--------------|-----|--------------------|-----|
| | | SD | %CV | SD | %CV |
| 1 | 20.14 | 0.96 | 4.8 | 0.60 | 3.0 |
| 2 | 23.79 | 8.11 | 5.6 | 0.35 | 1.5 |
| 3 | 37.54 | 0.72 | 3.0 | 1.36 | 3.6 |
| 4 | 144.18 | 0.84 | 2.2 | 4.56 | 3.2 |
| 5 | 281.57 | 6.17 | 2.2 | 17.85 | 6.4 |

b. *Linearity/assay reportable range:*

Linearity was evaluated based on CLSI EP6-A using one lot of reagent and one SPA_{plus} Analyzer. The study was performed by preparing two urine sample pools (high and low) that were intermixed to create a total of 12 samples that span from 7.785 – 394.662 mg/L and were each tested in replicates of 3. The cubic fit was significant in this analysis based on unweighted regression models. The mean test results were compared to the linear regression and the nonlinear cubic fit to evaluate non-linearity as a percentage value. The test results did not deviate from linearity by more than 3.7%. The results of the linear regression analysis were:

$$y = 0.99x - 0.11$$

$$r = 0.999$$

The linearity data provided support the sponsor's claims that the reportable range of this assay is 11 – 344 mg/L for human urine samples.

Antigen Excess:

A study was to support the claim that there was no hook effect with samples of albumin concentrations up to 8,000 mg/L. The study evaluated test results from high concentration samples that were diluted and found that samples up to and including 8,000 mg/L exhibited test results that were not impacted by antigen excess effects.

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

This kit is traceable to the reference standard European Reference Material ERM-DA470k. Kit calibrators are prepared using normal human serum and verified using a commercially available assay. The calibrators are prepared to target the following concentrations:

| Calibrator | Target value (mg/L) |
|-------------------|----------------------------|
| Cal 1 | 10.80 |
| Cal 2 | 21.50 |
| Cal 3 | 43.00 |
| Cal 4 | 86.00 |
| Cal 5 | 172.00 |
| Cal 6 | 344.00 |

The protocols and acceptance criteria for value assignment were reviewed and found acceptable.

Stability

Shelf Life Stability:

A real time stability study of the Human Microalbumin Kit was performed. The protocols and data were reviewed and found acceptable. The study supports a claim to a shelf life of 18 months, when stored at 2-8°C.

Open Vial Stability:

An Open Vial stability study for the Human Microalbumin Kit was performed. The protocols and acceptance criteria were reviewed and found acceptable. The study supports the claim that all components of the kit are stable 3 months after opening when stored at 2-8°C.

Reagent On-Board Stability:

On board stability study for the Human Microalbumin Kit protocols and data were reviewed and found acceptable. These studies support the claim that the on board stability for the antiserum and reaction buffer is 30 days.

d. *Detection limit:*

A Limit of Blank (LoB), Limit of Detection (LoD) and Limit of Quantitation (LoQ) study was performed based on CLSI EP17-A Protocols for Determination of Limits

of Detection and Limits of Quantitation and analyzed as described below.

To estimate LoB, five depleted urine samples were tested in replicates to yield 60 measurements with 1 kit lot and 1 analyzer. To estimate LoD, 5 normal human urine samples were diluted with saline to concentrations close to the bottom of the measuring range were tested 12 times over 5 days to yield 60 measurements per sample using 1 reagent kit lot on 1 analyzer.

The LoQ was estimated using the test results from the LoD measurements. The LoQ was defined using Total Error (TE) with the bias calculated in comparison to a legally marketed device. This assay exhibited 1.36 mg/L TE at the low end of the measuring range and supported the claimed LoQ.

The above studies support the following LoB, LoD and LoQ claims:

| Detections Limit Parameter | Value (mg/L) |
|----------------------------|--------------|
| LoB | 4.20 |
| LoD | 4.55 |
| LoQ | 11.00 |

The measuring range of the assay is 11 – 344 mg/L based on the LoQ and the linear range of the assay (see section M 1.b above).

e. Analytical specificity:

Interference studies were designed according to the CLSI EP7-A guideline. Three base pool samples with three clinically relevant albumin concentrations (20 mg/L: normal sample within the reference range, 30 mg/L: close to medical decision point and 55 mg/L: pathological sample) were spiked with possible interferents (10 drugs and 2 endogenous compounds). The % interference was calculated using the mean of the test sample and the mean of the negative control sample. Significant interference was defined as results $\geq 10\%$ of control. Results are summarized in the table below:

| Interferent | Highest Concentration Tested | Percent Interference | | |
|---------------------|------------------------------|----------------------|---------|---------|
| | | 20 mg/L | 30 mg/L | 55 mg/L |
| Hemoglobin | 250 mg/L | 7.01 | -4.84 | -7.48 |
| Bilirubin | 200 mg/L | 2.90 | -3.90 | 0.77 |
| Acetaminophen | 1324 μ mol/L | -6.75 | -2.37 | -3.67 |
| Ascorbic Acid | 342 μ mol/L | -5.37 | -3.33 | 3.75 |
| Furosemide | 90 μ mol/L | 5.44 | -5.20 | -1.27 |
| Ibuprofen | 2425 μ mol/L | 7.46 | 0.60 | -1.95 |
| Glybenclamide | 3.89 μ mol/L | 1.73 | 0.36 | 3.16 |
| Trichloromethiazide | 50mg/mL | 1.51 | 5.53 | 2.95 |
| Metformin HCL | 8 mg/L | -4.68 | -1.98 | -0.98 |
| Enalapril | 496.7 ng/mL | -2.22 | 0.18 | 6.22 |

| Interferent | Highest Concentration Tested | Percent Interference | | |
|-------------|------------------------------|----------------------|---------|---------|
| | | 20 mg/L | 30 mg/L | 55 mg/L |
| Losartan | 12.9 ng/mL | -1.37 | 3.32 | 7.44 |
| Simvastatin | 13.36 mg/mL | -0.24 | 0.81 | -4.91 |

Another interference study was conducted according to the CLSI EP7-A guideline. Two base pool samples with albumin concentrations targeting a low normal concentration (~22.5 mg/ml) and a high concentration sample (~200 mg/L: pathological sample) were spiked with possible interferents. The % interference was calculated using the mean of the test sample and the mean of the negative control sample. Significant interference was defined as results $\geq 10\%$ of control. The study is summarized in the table below:

| Interferent | Highest Concentration Tested | Percent Interference | |
|----------------|------------------------------|----------------------|----------|
| | | 22.5 mg/L | 200 mg/L |
| Calcium | 390 mg/L | -3.01 | 0.82 |
| Creatinine | 6000 mg/L | -4.59 | -1.17 |
| Glucose | 30000 mg/L | -7.43 | -3.61 |
| Sodium Citrate | 1000 mg/L | -1.32 | -2.08 |
| Ascorbic Acid | 2500 mg/L | -9.38 | 0.39 |
| Magnesium | 8000 mg/L | -4.07 | 0.18 |
| Acetaminophen | 6000 mg/L | -5.74 | -2.12 |
| Sodium oxalate | 600 mg/L | -4.69 | -3.11 |
| Urea | 50000 mg/L | 8.73 | -4.44 |
| Uric Acid | 200 mg/L | -.350 | -1.45 |
| Salicylic Acid | 1500 mg/L | -.293 | 2.22 |
| Metronidazole | 390 mg/L | >10 | >10 |
| Urobilinogen | 45 mg/L | 1.21 | 2.91 |

Metronidazole exhibited interference with test results and is included in the labeling as a limitation.

f. Assay cut-off:

Not applicable.

2. Comparison studies:

a. Method comparison with predicate device:

A method comparison study was performed using 90 human urine samples ranging from 12.0 mg/L to 332.0 mg/L. These samples were tested using the Human Microalbumin Kit using the SPA_{PLUS} and on the predicate device. The method comparison study was performed according to the CLSI EP9-A2 guideline. Samples were tested in singlicate and no spiked or diluted samples were included.

Passing Bablok regression analysis (n = 90) with 95% CI indicated in parentheses.

slope = 0.99 (0.96 – .99)

y-intercept = -0.1 (-0.87 to 0.62)

r = 0.982

In order to support that an auto- dilution of 1:10 by the SPA_{PLUS} will provide accurate test results in the range of 11 – 3440 mg/L, an additional method comparison study was provided using 121 human urine samples ranging from 12.0 mg/L to 2740 mg/L. Samples were tested in singlicate using the Human Microalbumin Kit using the SPA_{PLUS} and on the predicate device. The method comparison study was performed according to CLSI EP9-A2 guideline. Nine of the 121 samples were contrived (7.4%). Passing Bablok regression analysis (n = 121) with 95% CI indicated in parentheses.

slope = 0.97 (0.96 – .99)

y-intercept = 0.15 (-0.53 to 0.76)

r = 0.99

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):

Not applicable.

4. Clinical cut-off:

Not applicable.

5. Expected values/Reference range:

The expected values are included in the labeling and are taken from a literature reference¹.

| | 24 hr collection (mg/24 hr) | Timed collection (mg/min) | Spot Collection (µg/mg Creatinine) |
|-------------------------|--------------------------------|---------------------------------|---------------------------------------|
| Normal | < 30 | <20 | <30 |
| Microalbuminuria | 30 – 299 | 20 – 199 | 30 – 299 |
| Clinical albuminuria | ≥ 300 | ≥ 200 | ≥ 300 |

¹ American Diabetes Association. Diabetic Nephropathy. Diabetes Care 25: (Suppl. 1): S85-S89.

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.