510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:
   k071210
B. Purpose for Submission:
   Design changes (detection system with new platform software)
C. Measurand:
   Anti-SS-A, anti-SS-B, anti-Sm, anti-Sm/RNP, anti-dsDNA, anti-Scl-70, anti-Jo1, ribosome and centromere
D. Type of Test:
   Multiplex bead-based flow cytometric immunoassay
E. Applicant:
   Biomedical Diagnostics (bmd) S.A.
F. Proprietary and Established Names:
   FIDIS™ Connective 10*
G. Regulatory Information:
   1. Regulation section:
      21CFR§ 866.5100, Antinuclear Antibody Immunological Test System
   2. Classification:
      Class II
   1. Product code:
      LLL, Extractable Antinuclear Antibody, Antigen, and Control
      LKJ, Antinuclear Antibody, Antigen, Control
      LKO, Anti-RNP Antibody, Antigen, Control
      LKP, Anti-Sm Antibody, Antigen, and Control
      LSW, Anti-DNA Antibody, Antigen and Control
      LJM, Antinuclear Antibody (Enzyme Labeled), Antigen, Controls
      MQA, Anti-Ribosomal P Antibodies
   4. Panel:
      Immunology (82)
H. Intended Use:
   1. Intended use(s):
      The FIDIS ™ Connective 10* kit is a fluorescent immunoassay for the semi-quantitative simultaneous detection of 10 autoantibody specificities directed against double stranded DNA (dsDNA), SSA (60 kDA and 52 kDA), SSB, Sm, Sm/RNP, Scl-70, Jo-1 ribosome and centromere in human serum. (*Antibodies to dsDNA, SSA, SSB, Sm, Sm/RNP, Scl-70, Jo-1, ribosome and centromere can be reported using this assay).
   2. Indication(s) for use:
      The test system is used to screen serum samples and detect the presence of antinuclear antibodies associated with connective diseases systemic lupus erythematosus (SLE), Sjogren's syndrome, mixed connective tissue disease (MCTD), scleroderma, dermatomyositis, and CREST syndrome, in conjunction with clinical findings and other laboratory tests.
   3. Special conditions for use statement(s):

This device is for prescription use only.

4. Special instrument requirements:
   FIDIS™ Instrument (Luminex 200™ plus FIDIS™ MLX-Booster 2.2Software)
   CARIS™ (Optional diluting and dispensing device)

I. Device Description:

The device consists of the following: color-coded sets of microspheres (lyophilized). Each microsphere set is conjugated to one of the following antigens: dsDNA, SSa (60 kDA and 52 kDA), SSB, Sm, Sm/RNP, Scl-70, Jo-1, ribosome and centromere; calibrator (ready to use); positive control (to be diluted); negative control (to be diluted); goat anti-human IgG conjugate coupled phycoerythrin (ready to use) and washing buffer (ready to use).

J. Substantial Equivalence Information:

1. Predicate device name(s):
   FIDIS ™ Connective 10*
2. Predicate 510(k) number(s):
   k053653
3. Comparison with predicate:

Similarities		
Item	Device	Predicate
	Modified FIDIS™ Connective 10*	Initial FIDIS™ Connective 10*
Intended Use	Individual determination of IgG antibodies to dsDNA, SSA 60 kDA and 52 kDA, SSB, Sm, Sm/RNP, ScL-70, Jo-1, ribosome and centromere	Same
Assay type	Flow cytometer based	Same
Assay format	Multiplexed	Same
Solid phase capture	Color-coded microsphere	Same
Conjugate	Phycoerythin	Same
Sample type	Serum	Same
Type of test	Semi-quantitative	Same

Differences		
Item	Modified FIDIS™ Connective 10*	Initial FIDIS™ Connective 10*
Material supplied	Microplate without caps	Microplate with caps
Beads	Vial of color-coded microsphere set Lyophilized (sp 6mL)	6mL of vial of color-coded microsphere set ready to use
Sample Dilution	Sample dilution buffer ready to use	PBS-Tween concentrated
Wash buffer	Sample dilution buffer ready to use	PBS-Tween concentrated
Assay configuration	1 "reagent-blank" well 1 "negative control" well	1 "reagent-blank" well 1 "calibrator" well

Differences		
Item	Modified FIDIS™ Connective 10*	Initial FIDIS™ Connective 10*
	1 "positive control" well 2 "calibrator" wells Diluted sample wells	1 "negative control" well 1 "positive control" well Diluted sample wells A second calibrator well every 32 well series
Assay protocol	Final wash step (not optional)	Optional final wash step
Software	Booster Version 2.2	Booster Version 1.35
Detection	Based on Luminex 200	Based on Luminex 100

K. Standard/Guidance Document Referenced (if applicable):
CLSI C24A - Statistical Quality Control for Quantitative Measurement Procedures: Principles and Definitions
CLSI M29A – Protection of Laboratory Workers from Occupationally Acquired Infection
CLSI H18A –Procedures for the Handling and Processing of Blood Specimens

L. Test Principle:
FIDIS™ CONNECTIVE 10* is based on the use of distinct uniform size color-coded microspheres and a benchtop flow cytometer interfaced to a digital signal processing hardware and software. A red diode laser beam of the flow cytometer classifies each set of microspheres on the basis of its unique fluorescence intensity (red to orange) which allows to identify which analyte is being tested. At the same time, a green laser beam illuminates the external second molecule fluorescence to quantify the specific reaction related to each analyte.

Each antigen required for the assay is covalently coupled to an individual set of microspheres through its surface functional groups. The different antigen-coupled microspheres are mixed together to constitute the final microsphere reagent.
FIDIS™ CONNECTIVE 10* allows the detection of 10 autoantibody specificities: double stranded DNA (dsDNA), SSA 60kDa, SSA 52kDa, SSB, Sm, Sm/RNP, Scl70, Jo-1, Ribosomes and Centromeres. The microspheres are classified on the basis of their unique fluorescence intensity ratio that allows the identity of analyte being tested. Each dot in a white plot corresponds to an individual microsphere.

Seventy (70) µL of each sample are needed for each analysis. FIDIS™ Connective 10* assay was optimized by flow cytometry for the average binding capacity at the given dilution (1:200) from the median fluorescence value using 200 microspheres per parameter.

M. Performance Characteristics (if/when applicable):
1. Analytical performance:
   a. *Precision/Reproducibility:*
      To evaluate intra-assay and inter assay reproducibility, four samples covering the reportable range of the assay were analyzed on the modified FIDIS™ Connective 10*. For within-run, the four samples were assayed 6 times in one run and for between-run; the four samples were assayed 2 times per run for 6 runs. Additional study was performed for dsDNA on the low end of the measuring range. Results

were as follows:

Antigen	Within-run (10 tests in the same run)		Between-run (3 tests in 6 different runs)	
	Mean value	CV (%)	Mean value	CV (%)
dsDNA	496	5.9	508	10.5
	87	6.1	104	13.8
	63	8.9	63	6.7
	52	8.0	64	12.6
	14	8.2	14	10.8
	12	8.6	14	8.3
SSA 60 kDa and 52 kDa	209	2.1	202	5.4
	81	7.8	81	4.5
	64	5.3	64	5.5
	19	11.2	23	8.8
SSB	192	2.6	176	8.8
	74	8.0	76	7.0
	26	5.2	25	14.6
	19	2.5	18	10.9
Sm	306	3.2	303	6.8
	81	9.1	87	9.4
	66	8.6	63	10.8
	49	12.7	48	12.7
Sm/RNP	236	8.4	236	9.3
	133	7.9	139	7.6
	92	11.5	100	14.3
	75	6.2	70	9.5
Scl70	278	4.5	281	7.1
	162	5.4	153	9.2
	67	4.7	67	6.5
	5	5.9	6	13.6
Jo1	282	9.7	268	9.7
	100	3.0	92	9.4
	57	7.7	65	11.5
	9	12.8	9	12.2
Centromere	225	9.0	223	10.1
	94	3.6	92	6.7
	25	9.0	25	8.2
	22	6.7	21	8.5
Ribosome	154	6.9	152	10.1
	73	6.9	79	11.8
	50	6.4	53	8.4
	15	4.2	17	14.4

b. *Linearity/assay reportable range:*
   Linearity is not claimed for this assay.
c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*
   The dsDNA values in the calibrator are established using the WHO International Standard for anti-double stranded DNA (dsDNA), human code: WO/80. Other calibrator titers are expressed in arbitrary units per mL (AU/mL).
d. *Detection limit:*
   Not applicable
e. *Analytical specificity:*
   Interfering substances
   To evaluate the system for potential cross reactivity to other antibodies and interference from blood components, 30 samples were tested. High level of complement proteins were used but the specific kind of complement was not provided. A statement to avoid the use of abnormal concentration of these samples was added to the Limitations of the Procedure. The following results were obtained:

	Number of Positive Samples								
	dsDNA	SSA 60/52kD	SSB	Sm	Sm/RNP	Scl70	Jo1	Centromere	Ribosome
Cryoglobulinemia (2)									
Complement (7)	2	1	1	1	1				
IgG monoclonal Ig (1)									
IgM monoclonal Ig (5)									
Rheumatoid Factor (8)	1	2/2	1						
Blood Plasma (3)									
Hemolyzed sera (3)									
Anti-smooth muscle antibodies (1)									

f. *Assay cut-off:*
   Not applicable
2. Comparison studies:
   a. *Method comparison with predicate device:*
   The tables below show the comparison of serum samples (N=264) that were tested with the Modified FIDIS™ Connective 10* and the predicate device (Initial FIDIS™ Connective 10*). No information about age, gender, and clinical status was provided.
   - 194 positive samples for one or more parameters related to systemic autoimmune diseases
   - 70 negative samples
   All borderline results with the two devices were considered negative.

		Initial FIDIS™ Connective **10* dsDNA**		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **dsDNA**	Pos	45	1	46
	Neg	4	66	70
	Total	49	67	116

Positive % agreement: 91.84% (95% CI: 84.2% - 99.5%)

Negative % agreement: 98.51% (95% CI: 95.6% - 100%)
Overall % agreement: 95.69% (95% CI: 92.0% - 99.4%)

		Initial FIDIS™ Connective **10*** SSA 60/kDa		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **SSA 60/kDa**	Pos	48	1	49
	Neg	0	69	70
	Total	48	70	119

Positive % agreement: 100%
Negative % agreement: 98.57% (95% CI: 95.8%-100%)
Overall % agreement: 99.15% (95% CI: 97.52% - 100%)

		Initial FIDIS™ Connective **10*** SSA 52kDa		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **SSA 52kDa**	Pos	58	2	60
	Neg	3	64	67
	Total	61	66	127

Positive % agreement: 95.08% (95% CI: 89.7%, -100.0%)
Negative % agreement: 96.97% (95% CI: 92.8 – 100%)
Overall % agreement: 96.06% (95% CI: 92.7% -99.4%)

		Initial FIDIS™ Connective **10*** SSB		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **SSB**	Pos	23	2	25
	Neg	0	68	68
	Total	23	70	93

Positive % agreement: 100%
Negative % agreement: 97.14% (95%CI: 93.2% - 100%)
Overall % agreement: 97.85% (95% CI: 94.9% - 100%)

		Initial FIDIS™ Connective **10* Sm**		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **Sm**	Pos	23	2	25
	Neg	1	73	74
	Total	24	75	99

Positive % agreement: 95.83% (95% CI: 87.8% - 100%)
Negative % agreement: 97.33% (95%CI: 93.7% - 100%)
Overall % agreement: 96.97% (95% CI: 93.6%- 100%)

		Initial FIDIS™ Connective **10* Sm/RNP**		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **Sm/RNP**	Pos	31	2	33
	Neg	1	76	77
	Total	32	78	110

Positive % agreement: 96.88% (95% CI: 90.8% - 100%)
Negative % agreement: 97.44% (95%CI: 93.9% - 100%)
Overall % agreement: 97.27% (95% CI: 94.2% - 100%)

		Initial FIDIS™ Connective 10* Scl70		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **Scl70**	Pos	27	2	29
	Neg	1	68	69
	Total	28	70	98

Positive % agreement: 96.43% (95%CI: 89.2%-100%)
Negative % agreement: 97.14% (95%CI: 93.2% - 100%)
Overall % agreement: 96.94% (95% CI: 93.5% - 100%)

		Initial FIDIS™ Connective 10* Jo1		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **Jo1**	Pos	26	0	26
	Neg	1	69	70
	Total	27	69	96

Positive % agreement: 96.3% (95%CI: 89.2% - 100.0%
Negative % agreement: 100%
Overall % agreement: 98.96% (95% CI: 96.9% - 100%)

		Initial FIDIS™ Connective 10* Centromere		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **Centromere**	Pos	20	0	20
	Neg	0	69	69
	Total	20	69	89

Positive % agreement: 83.33% (95%CI: 68.4% - 98.2%)
Negative % agreement: 100%
Overall % agreement: 95.83% (95% CI: 91.8% - 99.8%)

		Initial FIDIS™ Connective 10* Ribosome		
		Pos	Neg	Total
Modified FIDIS™ Connective 10* **Ribosome**	Pos	18	0	18
	Neg	0	69	69
	Total	18	69	87

Positive % agreement: 100%
Negative % agreement: 100%
Overall % agreement: 100%

Comparison of the automated CARIS system and manual method
A comparison study between the manual method and the automated CARIS™ system was also performed. All borderline results with the two methods were considered negative.

dsDNA		Manual FIDIS		
		Pos	Neg	Total
Caris FIDIS	Pos	43	0	43
	Neg	0	69	69
	Total	43	69	112

Positive % agreement: 100.0%
Negative % agreement: 100.0%
Overall % agreement: 100.0%

SSA (60kDa)		Manual FIDIS		
		Pos	Neg	Total
Caris FIDIS	Pos	42	0	42
	Neg	0	68	68
	Total	42	68	110

Positive % agreement: 100.0%
Negative % agreement: 100.0%
Overall % agreement: 100.0%

SSA (52kDa)		Manual FIDIS		
		Pos	Neg	Total
Caris FIDIS	Pos	52	1	53
	Neg	0	65	65
	Total	52	66	118

Positive % agreement: 100.0%
Negative % agreement: 98.5%
Overall % agreement: 99.2%

SSB		Manual FIDIS		
		Pos	Neg	Total
Caris FIDIS	Pos	25	0	25
	Neg	0	67	67
	Total	25	67	92

Positive % agreement: 100.0%
Negative % agreement: 100.0%
Overall % agreement: 100.0%

Sm		Manual FIDIS		
		Pos	Neg	Total
Caris FIDIS	Pos	24	3	27
	Neg	0	69	69
	Total	24	72	96

Positive % agreement: 100.0%
Negative % agreement: 95.8%
Overall % agreement: 96.9%

		Manual FIDIS		
Sm/RNP		Pos	Neg	Total
Caris FIDIS	Pos	32	3	35
	Neg	0	68	68
	Total	32	71	103

Positive % agreement: 100.0%
Negative % agreement: 95.8%
Overall % agreement: 97.1%

		Manual FIDIS		
Scl70		Pos	Neg	Total
Caris FIDIS	Pos	29	0	29
	Neg	0	68	68
	Total	29	68	97

Positive % agreement: 100.0%
Negative % agreement: 100.0%
Overall % agreement: 100.0%

		Manual FIDIS		
Jo1		Pos	Neg	Total
Caris FIDIS	Pos	26	0	26
	Neg	0	69	69
	Total	26	69	112

Positive % agreement: 100.0%
Negative % agreement: 100.0%
Overall % agreement: 100.0%

		Manual FIDIS		
Centromere		Pos	Neg	Total
Caris FIDIS	Pos	20	4	24
	Neg	0	69	69
	Total	20	73	93

Positive % agreement: 100.0%
Negative % agreement: 94.5%
Overall % agreement: 95.7%

		Manual FIDIS		
Ribosome		Pos	Neg	Total
Caris FIDIS	Pos	8	0	8
	Neg	0	68	68
	Total	8	68	78

Positive % agreement: 100.0%
Negative % agreement: 100.0%
Overall % agreement: 100.0%

       b. Matrix comparison:
          Serum is the only recommended matrix.
3. Clinical studies:
       a. Clinical Sensitivity:
          Not provided
       b. Clinical specificity:
          Not provided
       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 reported expected ranges were estimated from 2 populations:
   • 50 samples from blood donors
   • 48 samples selected from their potential biological interferences and according to WHO standard for dsDNA specificity

Arbitrary units (AU/mL)	<30 AU/mL	30-40 AU/mL	>40 AU/mL
International units (IU/mL) For dsDNA	<30 IU/mL	30-40 IU/mL	>40 IU/mL
Interpretation	Negative	Equivocal[1]	Positive

The negative thresholds (30 AU/mL or 30 IU/mL) correspond to the 97.9^{th} percentile for dsDNA, SSA, Sm/RNP; 99.0% for centromere and ribosome, and 100% for SSB, Sm, Scl70 and Jo1 for the populations studied.

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.