510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:
K051144

B. Purpose for Submission:
New device.

C. Measurand:
Acetylcholine receptor autoantibodies.

D. Type of Test:
Semi-quantitative, Radio receptor assay

E. Applicant:
IBL-Hamburg GmbH

F. Proprietary and Established Names:
Acetylcholine Receptor Ab (ARAb) RRA

G. Regulatory Information:

1. Regulation section:
   21 CFR § 866.5660, Multiple Autoantibodies Immunological Test System
2. Classification:
   Class II
3. Product code:
   NST, Autoantibodies, Acetylcholine Receptor, Acetylcholine Blocking and Non-Blocking.
4. Panel:
   Immunology (82)

H. Intended Use:

1. Intended use(s):
   The IBL Acetylcholine Receptor Antibody (ARAb) Radio Receptor Assay is for semi-quantitative determination of autoantibodies against the acetylcholine receptor in human serum and plasma. The ARAb assay kit is useful as an aid in the differential diagnosis of Myasthenia Gravis (MG).
2. Indication(s) for use:
   Same as intended use.
3. Special conditions for use statement(s):
   The device is for prescription use only.
4. Special instrument requirements:
   Gamma counter for I^{125}.

I. Device Description:

The IBL Acetylcholine Receptor Antibody (ARAb) Assay Kit consists of:

1. Lyophilized I^{125} Acetylcholine Receptor from human muscle and labeled with I^{125} alpha-bungarotoxin.
2. I^{125} ARAb Tracer (lyophilized)
3. IgG Antiserum contains anti-human IgG, 0.1% NaN_3, ready-to-use.
4. Positive control contains human serum, antibodies to acetylcholine receptors-ready to use.

5. Standard A-F 0; 0.2; 0.5; 1.2; 3.0; 8.0nmol/L- ready to use.
   Standard A = sample diluent- normal human serum
6. Positive control, ready-to-use.
7. Wash buffer –PBS with 0.01% Triton X100. 0.1% NaN_3- ready to use.
8. Buffer for reconstitution of tracer, contains PBS,0.5 Triton and 0.005% NaN_3

J. Substantial Equivalence Information:

The sponsor claimed the new device is substantially equivalent to the Kronus Acetylcholine Receptor antibody Kit. The intended use and indications for use and methodology are the same. There is clinical data to support the claim.

1. Predicate device name(s):
   Kronus I^{125} Acetylcholine Receptor Antibody Kit
2. Predicate 510(k) number(s):
   k042248
3. Comparison with predicate:

Similarities		
Item	**Predicate**	**New device**
Device Name	Kronus Acetylcholine Receptor Antibody (AChRAb) Assay Kit	IBL Acetlylcholine receptor antibody assay (ARAb RRA)
Intended use	Detection of autoantibodies to specific receptor	Same
Test Principle	Radioreceptor immunoassay	Same
Test matrix	Serum	Serum and plasma
Test Platform	Receptors bind to serum antibodies in a liquid phase (test tube), the antibody-antigen complexes are precipitated and radioactivity measured	Same
Detection instrument	Gamma counter	Same
Differences		
Expected values Negative/cut-off Equivocal Positive	Serum< 0.2nmol/L ≤ 0.2nmol/L 0.21-049 nmol/L ≥ 0.5 nmol/L	Serum0.029 ± 0.018 nmol/L Plasma 0.023± 0.013 nmol/L < 0.25 nmol/L 0.25-0.4 nmol/L > 0.4 nmol/L
Detection limit	0.26 nmol/L	0.01nmol/L
Assay range	0.3-7.0 nmol/L	0.25-1.5 nmol/L

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

None referenced.

L. Test Principle:

Patient specimens and reference reagents are incubated at 18-25 °C for 2 hours with adult acetylcholine receptors (ARAb) labeled with I^{125} –alpha-bungarotoxin. The

resulting bound complexes of labeled ARAb and autoantibody are then immunoprecipitated with anti-human IgG. After centrifugation, the supernatant is aspirated and the pellet containing labeled ARAb/autoantibody-bound complex is counted in a gamma counter for 1 min. Counts are directly proportional to the amount of autoantibody present and results are expressed as the concentration of alpha-bungarotoxin bound (nmoles/L toxin bound). A standard curve is generated to calculate the values. All samples with a mean cpm above the cut off are considered positive.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Intra-assay precision was determined by analyzing 22 replicates of 7 control samples. The following table summarizes the results.

Sample	N	Mean ARAb (nmol/L)	SD (nmol/L)	% CV
1	22	0.19	0.01	5.1
2	22	0. 41	0.01	3.1
3	22	0. 75	0.02	2.8
4	22	1.55	0.06	3.8
5	22	2.62	0.17	6.3
6	22	4.57	0.39	8.5
7	22	5.72	0.65	11.3

Inter-assay variation was assessed by analyzing 7samples and 15 assay runs. The assays were performed in different labs with 3 different technicians using one kit lot. The mean coefficent of variation was found to be 70 % (range 2.8-13.1%). Results are shown in the table below.

Serum sample	Mean (nmol/L)	SD	CV %	N
1	0.21	0.01	5.3	14
2	0.40	0.01	2.8	15
3	0. 75	0.02	3.0	15
4	1.61	0.07	4.3	15
5	2.63	0.20	7.7	15
6	4.72	0. 59	12.4	15
7	6.10	0.80	13.1	15

b. Linearity/assay reportable range:

Four serum samples with different acetylcholine receptor antibody levels were diluted with Zero calibrator and Acetylcholine receptor antibody content, in ratios 1:2, 1:4, 1:8, 1:16, 1:32 and 1:64

Sample	Dilution	Measured (nmol/L)	Recovery (%)	Sample	Dilution	Measured (nmol/L)	Recovery (%)
1	-	7.038	100	3	-	2.711	100
	1:2	3.927	112		1:2	1.164	86
	!:4	1: 833	104		1:4	0.565	83
	1:8	0.924	105		1:8	0.263	78
	1:16	0.467	106		1:16	0.134	79
	1:32	0. 219	100		1:32	0.076	90
	1:64	0. 122	111		1:6	0.057	135
2	-	4.926	100	4	-	1.624	100
	1:2	2.568	104		1:2	0.808	100
	1:4	1.200	97		1:4	0.416	102
	1:8	0.553	90		1:8	0.205	101
	1:16	0.269	87		1:16	0.108	106
	1:32	0.124	81		1:32	0.048	95
	1:64	0.075	97		1:64	0.032	126

Recovery: Increasing amounts of acetylcholine receptor antibodies were added to serum samples with various initial acetylcholine receptor antibody concentrations. Each sample was assayed in duplicate in one run. % recovery rates were calculated. Mean recovery was 106% ± 19% (range 87-153%).

Sample	Endogenous ARAb (nmol/L)	Added ARAb (nmol/L)	Measured ARAb (nmol/L)	Recovery (%)
1	00	0.22	0.20	91
	00	0.60	0.52	87
	00	1.80	2.11	117
2	0.21	0.22	0.43	100
	0.21	0.60	0.87	107
	0.21	1.80	3.07	153
3	0.48	0..22	0.66	94
	0.48	0.60	0.97	90
	0.48	1.80	2.53	111

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*
No reference material available.

d. *Detection limit:*
The lower detection limit is 0.01nmol/L.

e. *Analytical specificity:*
Cross reactivity with autoantibodies potentially interfering with ARAb RRA was assessed. Nine patient serum samples containing autoantibodies (anti-Sm, -RNP, -RO (SSA), La (SS-B), -dsDNA,-RF, and ANA) were tested. No cross reactivities were found with any of the antibodies. In addition, hemoglobin (0.25 and 4 mg/mL), bilirubin (0.03 and 0.5 mg/mL and triglycerides (0.2 to 30mg/mL) were evaluated for interference. All test

results are available in the submission. Results showed no significant interference at the concentrations tested.

f. Assay cut-off:

One hundred and thirty serum and EDTA plasma samples from blood donors were assayed. Range of values were between 0.013 and 0116 nmol/L for serum and 0.013 and 0.086 nmol/L for plasma The upper limit of reference range is at 0.110 nmol/L for serum and 0.081 nmol/L for plasma. Cut off is set at 0.25 nmol/L. Antibody titers greater than 0.4 nmol/L are indicative of Myasthenia Gravis (positive). Values between 0.25 and 0.4 nmol/L is considered as equivocal

2. Comparison studies:

a. Method comparison with predicate device:

Fifty-three patient samples consisted of 38 Myasthenia Gravis positive samples and 15 Myasthenia Gravis negative samples were tested with the Kronus RIA test and IBL RRA test. Equivocal results are considered positive. Comparative results are summarized below:

		KRONUS			
		+	Equivocal	-	Total
IBL	+	37	0	0	37
	Equivocal	2	0	0	2
	-	0	2	12	14
	Total	39	2	12	53

% Positive agreement = 95.1% (39/41) (95% CI: 83.5% to 99.4%)
% Negative agreement = 100% (12/12) (95% CI: 73.5% to 100%)
% Total agreement = 96.2% (51/53)

b. Matrix comparison:

Seven patient samples, serum and EDTA plasma, comparison showed no significant difference. A correlation coefficent of linear regression was r = 0.984. Regression line was calculated (plasma) =0.931 (serum) + 0.062.

3. Clinical studies:

a. Clinical sensitivity and specificity:

The same samples used in the comparison study were also used determine the clinical sensitivity and specificity of the IBL RRA and the Kronus RIA. Results are shown in tables below.

		Myasthenia Gravis		
		+	-	Total
IBL	+	38	1	39
	-	0	14	14
	Total	38	15	53

Sensitivity = 100% (95% CI: 90.8% to 100%)
Specificity = 93.3% (95% CI: 68.1% to 99.8%)

		Myasthenia Gravis		
		+	-	Total
Kronus	+	38	3	41
	-	0	12	12
	Total	38	15	53

Sensitivity =100% (95% CI: 90.8 to 100%)
Specificity = 80% (95% CI: 51.9% to 95.7%)

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

4. Clinical cut-off**:**
See Assay cut-off.
5. Expected values/Reference range:
The expected value in normal population is less than 0.013 nmol/L.

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.