

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

A. 510(k) Number:

k042982

B. Purpose for Submission:

New Device

C. Measurand:

Myoglobin

D. Type of Test:

Quantitative Particle Enhanced Immunoturbidimetric assay

E. Applicant:

Instrumentation Laboratory Company

F. Proprietary and Established Names:

Quantex Myoglobin

Quantex Ferritin/Myoglobin Controls I/II

Quantex Myoglobin Standard Multipoint

G. Regulatory Information:

1. Regulation section:

21 CFR 866.5680 – Myoglobin Immunological Test System

21 CFR 862.1150 – Calibrator

21 CFR 862.1660 – Quality Control Material

2. Classification:

Class II, II, and I, respectively

3. Product code:

DDR, JIS, JJX

4. Panel:

Immunology (82); Chemistry (75)

H. Intended Use:

1. Intended use(s):

See Indications for Use statement below

2. Indication(s) for use:

Quantex Myoglobin is intended as a latex particle enhanced immunoturbidimetric assay for the quantitative determination of myoglobin concentration in human serum or plasma (EDTA or Lithium Heparin) on Clinical Chemistry Systems as an aid in the diagnosis of myocardial infarction. For *in vitro* diagnostic use.

Quantex Ferritin / Myoglobin controls I/II are intended for use in monitoring the quality control of results obtained with the quantex Myoglobin reagents by turbidimetry. (NOTE: These controls were previously FDA cleared for use with quantex Ferritin, reference K040879.) For *in vitro* diagnostic use.

Quantex Myoglobin standard multipoint is intended for use in establishing the calibration curve for the quantex Myoglobin reagents by turbidimetry. For *in vitro* diagnostic use.

3. Special conditions for use statement(s):

For Prescription Use Only

4. Special instrument requirements:

Instrument Laboratory Company provides an instrument sheet for the ILAB600.

I. Device Description:

The Instrumentation Laboratory's Quantex Myoglobin reagent kit is designed to measure myoglobin concentration in serum or plasma (EDTA or Lithium Heparin) using the ILAB600 or other clinical chemistry systems. The device consists of two reagents: buffer reagent and latex reagent. The latex reagent is comprised of polystyrene latex particles coated with rabbit IgG anti-human myoglobin. The myoglobin in a sample is directly proportional to the degree of transmitted light caused by the aggregates. The Quantex Myoglobin Standard Multipoint is a 5-level glycine buffer containing human myoglobin. The Quantex

Ferritin/Myoglobin controls I/II is a lyophilized solution of buffer with human myoglobin. The Quantex Myoglobin Standard multipoint and the Quantex Ferritin/ Myoglobin controls I/II are sold separately. All human source material used in the preparation of this product was found to be negative for the presence of HIV-1/2 and HCV antibodies, as well as for the hepatitis B surface antigen, using a FDA approved method.

J. Substantial Equivalence Information:

1. Predicate device name(s):

N Latex Myoglobin

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

k902154

3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	Quantitative determination of myoglobin concentration in human serum or plasma (EDTA or Lithium Heparin) on Clinical Chemistry Systems as an aid in the diagnosis of myocardial infarction.	Same
Sample Type	Serum or Plasma (EDTA or Lithium Heparin)	Serum or Plasma
Storage Conditions	2-8°C	Same

Differences		
Item	Device	Predicate
Method	Particle Enhanced Immunoturbidimetry	Particle Enhanced Nephelometry
Calibrators	Glycine buffer containing human myoglobin. (5 levels)	Lyophilized pool of human sera supplemented with human myoglobin. (3 levels)
Controls	Lyophilized solution of buffer with human myoglobin. (2 levels)	Lyophilized pool of human sera supplemented with human myoglobin. (3 levels)

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

NCCLS EP5-A: Method Comparison of Precision Performance of Clinical Chemistry Devices; Approved Guideline.

L. Test Principle:

Quantex Myoglobin is a latex particle enhanced immunoturbidimetric assay to quantify myoglobin in human serum or plasma (EDTA or Lithium Heparin). Myoglobin present in a sample reacts with rabbit IgG antihuman myoglobin in the latex buffer and also with a reaction buffer. These set of reactions initiates a clear agglutination. The myoglobin in the sample is proportional to the degree of agglutination and is measured by the decrease of transmitted light caused by the aggregates.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. *Precision/Reproducibility:*

Precision was assessed and calculated according to NCCLS EP5-A. The study was performed on an ILab 600 using the quantex Ferritin/Myoglobin controls level I and II. Two controls (low and high) were assayed in duplicate twice a day for 20 days (number of observations=80). The results are summarized below (units =ng/mL).

Material	N	Mean (ng/mL)	Within Run %CV	Between Run %CV	Total %CV
Low Control (I)	80	71.4	1.1	1.0	1.9
High Control (II)	80	228.6	1.3	1.8	2.3

b. *Linearity/assay reportable range:*

The sponsor identifies two reportable ranges:

- 1) ILab 600 that has an automatic rerun capability of 10 to 5000 ng/mL and
- 2) ILab 600 without a rerun capability of 10 to 500 ng/mL.

The ILab 600 automatically reruns samples with results above the upper limit of 500 ng/mL using a specific dilution (1:10) of the sample volume. The instrument then automatically recalculates the new sample result. The sponsor instructs users of analyzers with automatic rerun capability to dilute samples that produce myoglobin concentration values that are greater than 5000 ng/mL to dilute the sample 1:100 with saline, re-assay and to multiply the result by the dilution factor.

These ranges were tested using 5 serial dilutions of the highest level of the quantex Myoglobin standard multipoint (500 ng/mL). Each dilution was assayed with 3 lots of quantex reagent in 5 replicates to calculate the inaccuracy. Inaccuracy acceptable criteria is +/- 15%. Inaccuracy was calculated as

$$\text{Inaccuracy} = \frac{(\text{Reported value} - \text{Expected Value})}{(\text{Expected Value})} \times 100.$$

The reported values were compared to the expected values and the linearity correlation for the 3 lots was obtained.

Lot 1 $Y = 0.996x + 15.18$ $R^2 = 0.9982$

Lot 2 $Y = 0.999X + 15.09$ $R^2 = 0.9984$

Lot 3 $Y = 0.993X + 15.75$ $R^2 = 0.9984$

To evaluate the linearity after a sample has been diluted, a dilution study was conducted on the ILab 600. Two levels of serially diluted serum samples with saline were analyzed with one lot of the quantex Myoglobin reagent. The reported mean was calculated from the pooled results. The R^2 for the two samples were 0.993 and 0.999.

To analyze the hook effect using the Quantex Myoglobin, a study was conducted by the sponsor using a prozone control that has a myoglobin concentration that is >40,000 ng/mL. The prozone control was serially diluted to form 8 concentrations. The 8 samples were run in 5 replicates with 3 lots of the Quantex Myoglobin reagents. No prozone (high dose hook) effect was detected up to 47637 ng/mL since the reported results were above the linear range. See chart below.

Expected ng/mL	Reported Mean ng/mL		
	Lot 1 With Rerun	Lot 2 With Rerun	Lot 3 With Rerun
47637	10564.3	13015.3	***
23819	10095.8	11467.3	10541.0
11909	8018.0	8391.8	8125.3
5955	5138.5	5180.0	5127.5
2977	2738.0	2745.0	2700.0
1489	1356.5	1361.8	1368.3
744	683.8	694.8	689.3
372	372.5	372.8	370.3

***indicated that there is no result for the ILab 600 and appears when there is an absorbance value greater than the absorbance limit value set for the Myoglobin test.

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

Value assignments were determined in multiple runs on the ILab Clinical Chemistry Systems using specific lots of reagents and against a Calibration House standard that is traceable to a commercially available method.

The Quantex Ferritin/ Myoglobin control I/II are obtained from an external source (Denka Seiken). The control value range is assigned at Biokit by comparison to the Myoglobin Calibration House Standard.

On-board stability testing was conducted on the ILAB instrument and the results supported the claim of 2 months at 15°C. The Quantex Myoglobin (both Latex reagent and Reaction Buffer) were tested on day zero with 4 levels of Quantex Myoglobin standard multipoint, quantex controls (low and high), prozone control and a reagent blank in duplicate at multiple time periods.

Calibration stability testing was performed using an ILab Instrument with the Quantex Myoglobin reagents. The five levels of calibrators, and the prozone controls (1/2 diluted to 12800 ng/mL) were analyzed as samples with the quantex Myoglobin. The acceptance criteria were identified and supported the calibration stability of 2 months.

The stability of the reconstituted controls was tested in both the low and the high controls at 2-8°C. Two lots of the Quantex Ferritin/Myoglobin Controls I/II were reconstituted at 2-8°C and analyzed singly. The acceptance criteria were identified and the results supported the 14 days stability at 2-8°C.

d. *Detection limit:*

To calculate detection limit, saline was analyzed in runs of 10 replicates on three different lots of Quantex Myoglobin reagents. The mean value + 3 standard deviations were calculated for each of the three lots. The largest of the three results was used to establish the detection limits. Results are summarized in the table below.

	Lot 1	Lot 2	Lot 3
Mean (ng/mL)	1.04	0.74	0.37
SD	0.527	0.707	0.301
Detection Limit	2.622	2.862	1.283

The results support the claim for a detection limit = 3 ng/mL.

e. *Analytical specificity:*

Interference testing was performed by spiking levels of each interferent into two levels of pooled serum and comparing the results against the unspiked sample results. All samples were tested in triplicate with a single lot of Myoglobin Reagent. Acceptance criterion is recovery of +/- 15% of the unspiked sample result.

There was no interference (<10%) from the following substances.

- Lipemia up to a sample absorbance of 16.6 AU/cm at 660 nm,
- Triglycerides up to concentrations of 1300 mg/dL,
- Bilirubin up to concentrations of 20 mg/dL,
- Hemoglobin up to concentrations of 500 mg/dL,
- Iodine containing contrast media (Omnipaque) up to concentrations of 25 g/L and
- Streptokinase up to concentrations of 1500 IU/mL.

There was no significant cross reactions (<12%) from the following substances.

- Rheumatoid factor up to concentrations of 800 IU/mL,
- Human IgG up to concentrations of 60 g/L,
- Human albumin up to concentrations of 60 g/L,
- Human haptoglobin up to concentrations of 5 g/L and
- Human hemopepsin up to concentrations of 4 g/L.

Fresh serum or plasma (EDTA or Lithium Heparin) samples only are to be used. Other sample types are not validated for use with this assay. Heat inactivated serum samples are not to be used.

f. *Assay cut-off:*

Functional sensitivity testing was performed on the ILab 600 using 5 saline dilutions of the 62.5 ng/mL Quantex Myoglobin standard multipoint. The samples were run with 3 different lots of the Quantex Myoglobin reagents in replicates of 10. The sponsor acceptance criterion is a CV less than 15%. The results supported the products claim for functional sensitivity of 10 ng/mL.

2. Comparison studies:

a. *Method comparison with predicate device:*

67 patient serum samples ranging from 15.6 to 2070 ng/mL were analyzed using Quantex Myoglobin on an ILab 600 and with the Dade Behring's N Latex Myoglobin (predicate device) on a Behring Nephelometer System. No

artificially prepared samples were used in this study. 60 samples were from an emergency room and 7 were from blood bank donors. The acceptance criteria are a slope of 0.80 to 1.20 and a correlation coefficient greater than .980.

The following results were obtained:

$$\text{Quantex Myoglobin} = 0.9909x - 2.3865$$

The equation has a $R^2 = 0.997$ and correlation coefficient = 0.9985.

b. *Matrix comparison:*

Plasma to serum correlation was conducted with 2 sets of paired samples : 10 EDTA plasma samples paired to serum and 10 Lithium Heparin plasma samples paired to serum. Each paired sample was spiked with purified myoglobin and retested (n=20). The samples were analyzed in duplicate with one lot of Quantex Reagents. The acceptance criteria to show sample type equivalence is a recovery of +/- 15% of the serum sample result. The recoveries ranged from 94.4% to 106.2%. A plot of serum values (ng/mL) versus EDTA plasma (ng/mL) yielded a R^2 of 0.9994. A plot of Serum values (ng/mL) versus Lithium Heparin Plasma values (ng/mL) yielded a R^2 of 0.9994.

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 values for the upper and lower limit for apparently healthy individuals (175 males and 181 females) was calculated based on 360 samples from blood bank

donors. Four samples were removed as far outliers (1 inter-quartile range above the mean). The table below shows the breakdown of the data by gender and the non-parametric reference interval calculated as recommended by the International Federation of Clinical Chemistry (IFCC).

	Women		Men	
	95% Limit	90 % CI	95% Limit	90% CI
Lower	12.8	7.5-13.5	16.2	13.5-17.0
Upper	42.9	40.0-54.5	76.3	58.0-96.0
Mean	22.7		31.9	
N	181		175	

The data supports the reported upper limit for the reference interval that is stated in the package insert as 43 ng/mL for women and 76 ng/mL for men.

The sponsor cautions that each laboratory should establish its own reference range.

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.