510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K253358

B Applicant

Diazyme Laboratories, Inc.

C Proprietary and Established Names

Diazyme Human Kappa (κ) Free Light Chain Assay
Diazyme Human Lambda (λ) Free Light Chain Assay

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
DFH, DEH	Class II	21 CFR 866.5550 - Immunoglobulin (Light Chain Specific) Immunological Test System	IM - Immunology

II Submission/Device Overview:

A Purpose for Submission:

Modification of a previously cleared device: Addition of intended use as an aid in the evaluation of Monoclonal Gammopathy of Undetermined Significance (MGUS).

B Measurand:

Kappa (κ) Free Light Chain (FLC)
Lambda (λ) Free Light Chain (FLC)

C Type of Test:

Latex particle enhanced immunoturbidimetric, quantitative

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The Diazyme Human Kappa (κ) Free Light Chain Assay is intended as a latex particle enhanced immunoturbidimetric assay for the quantitative determination of Kappa Free Light Chain (FLC) concentration in serum on validated analyzers. The measurement of Kappa FLC in conjunction with Lambda FLC aids in the diagnosis and monitoring of multiple myeloma and aids in the evaluation of monoclonal gammopathy of undetermined significance (MGUS), in conjunction with other laboratory and clinical findings. For *in-vitro* diagnostic use only.

The Diazyme Human Lambda (λ) Free Light Chain Assay is intended as a latex particle enhanced immunoturbidimetric assay for the quantitative determination of Lambda Free Light Chain (FLC) concentration in serum on validated analyzers. The measurement of Lambda FLC in conjunction with Kappa FLC aids in the diagnosis and monitoring of multiple myeloma and aids in the evaluation of monoclonal gammopathy of undetermined significance (MGUS), in conjunction with other laboratory and clinical findings. For *in-vitro* diagnostic use only.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

The following statements are in the package insert of the Assays:

Warning: Kappa FLC result for a given specimen determined with assays and/or instrument platforms from different manufacturers can vary due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the FLC assay used. Values obtained with different assay methods cannot be used interchangeably. If, in the course of serially monitoring a patient, the assay method used for determining the Kappa FLC levels is changed, additional sequential testing should be carried out. Prior to changing assays, the laboratory should confirm baseline values for patients being serially monitored.

Warning: Lambda FLC results for a given specimen determined with assays and/or instrument platforms from different manufacturers can vary due to differences in assay methods and reagent specificity. The results reported by the laboratory to the physician must include the identity of the FLC assay used. Values obtained with different assay methods cannot be used interchangeably. If, in the course of serially monitoring a patient, the assay method used for determining the Lambda FLC levels is changed, additional sequential testing should be carried out. Prior to changing assays, the laboratory should confirm baseline values for patients being serially monitored.

Evaluation of monoclonal gammopathy of undetermined significance (MGUS):

- The performance has not been sufficiently studied in Light Chain MGUS patients.

- Patients with renal disease or inflammation may have elevated levels of kappa and lambda free light chains (FLC).
- The performance has not been fully evaluated in all races/ethnicities in the intended use population.

D Special Instrument Requirements:

Roche cobas c 501 analyzer

IV Device/System Characteristics:

A Device Description:

No modification is made to the kit components for the Diazyme Human Kappa (κ) and Lambda (λ) Free Light Chain Assays cleared in K153394.

The Diazyme Human Kappa (κ) Free Light Chain Assay is comprised of the following reagents:

- Reagent R1: Tris buffer solution
- Reagent R2: Carboxyl Polystyrene particles coated with polyclonal rabbit and goat anti-Kappa FLC antibodies
- Calibrators: A five-level set in liquid form (5 x 1 mL)

The Diazyme Human Lambda (λ) Free Light Chain Assay is comprised of the following reagents:

- Reagent R1: Tris buffer solution
- Reagent R2: Carboxyl Polystyrene particles coated with polyclonal rabbit anti-Lambda FLC antibodies
- Calibrators: A five-level set in liquid form (5 x 1 mL)

The controls are required materials but not provided for both assays. For each assay, the control set includes two levels of controls in serum based liquid form.

B Principle of Operation:

No modification is made to the principle of operation for the Diazyme Human Kappa (κ) Free Light Chain Assay or the Diazyme Human Lambda (λ) Free Light Chain Assay.

The assays are based on a latex enhanced immunoturbidimetric assay. Kappa FLC or Lambda FLC in the sample binds to specific anti-Kappa FLC or anti-Lambda FLC antibody, which is coated on latex particles, and causes agglutination. The degree of the turbidity caused by agglutination can be measured optically and is proportional to the amount of Kappa FLC or Lambda FLC in the sample. The instrument calculates the Kappa FLC or Lambda FLC concentration by interpolation of obtained signal of a 6-point calibration curve prepared from calibrators of known concentrations.

V Substantial Equivalence Information:

A Predicate Device Name(s):

Diazyme Human Kappa (κ) Free Light Chain Assay
Diazyme Human Lambda (λ) Free Light Chain Assay

B Predicate 510(k) Number(s):

K211648

C Comparison with Predicate(s):

Device & Predicate Device(s):	**K253358 (Candidate Device)**	**K211648 (Predicate Device)**
Device Trade Name	Diazyme Human Kappa (κ) Free Light Chain Assay Diazyme Human Lambda (λ) Free Light Chain Assay	Diazyme Human Kappa (κ) Free Light Chain Assay Diazyme Human Lambda (λ) Free Light Chain Assay
General Device Characteristic Similarities		
Analyte	Kappa FLC, Lambda FLC	Same
Assay Type	Quantitative	Same
Test Method	Turbidimetry	Same
Specimen Type	Serum	Same
Traceability	Internal Reference preparation	Same
Units	mg/L	Same
Detection Antibody	Polyclonal anti-human Kappa/anti-human Lambda FLC Ig fraction coated on polystyrene latex microparticles	Same
Analytical Measuring Interval	Kappa: 2.9–150 mg/L Lambda: 3.5–200 mg/L	Same
Reference Interval	Kappa: 2.4–20.7 mg/L Lambda: 4.2–27.7 mg/L Ratio: 0.22–1.74	Same
General Device Characteristic Differences		
Intended Use/ Indications for Use	The Diazyme Human Kappa (κ) Free Light Chain Assay is intended as a latex particle enhanced immunoturbidimetric assay for the quantitative determination of Kappa Free Light Chain (FLC) concentration in serum on validated analyzers. The measurement of Kappa FLC in	The Diazyme Human Kappa (κ) Free Light Chain Assay is intended as a latex particle enhanced immunoturbidimetric assay for the quantitative determination of Kappa Free Light Chain (FLC) concentration in serum on validated analyzers.

Device & Predicate Device(s):	K253358 (Candidate Device)	K211648 (Predicate Device)
	conjunction with Lambda FLC aids in the diagnosis and monitoring of multiple myeloma and aids in the evaluation of monoclonal gammopathy of undetermined significance (MGUS) in conjunction with other laboratory and clinical findings. For *in-vitro* diagnostic use only. The Diazyme Human Lambda (λ) Free Light Chain Assay is intended as a latex particle enhanced immunoturbidimetric assay for the quantitative determination of Lambda Free Light Chain (FLC) concentration in serum on validated analyzers. The measurement of Lambda FLC in conjunction with Kappa FLC aids in the diagnosis and monitoring of multiple myeloma and aids in the evaluation of monoclonal gammopathy of undetermined significance (MGUS) in conjunction with other laboratory and clinical findings. For *in-vitro* diagnostic use only.	The measurement of Kappa FLC in conjunction with Lambda FLC aids in the diagnosis and monitoring of multiple myeloma in conjunction with other laboratory and clinical findings. For *in-vitro* diagnostic use only. The Diazyme Human Lambda (λ) Free Light Chain Assay is intended as a latex particle enhanced immunoturbidimetric assay for the quantitative determination of Lambda Free Light Chain (FLC) concentration in serum on validated analyzers. The measurement of Lambda FLC in conjunction with Kappa FLC aids in the diagnosis and monitoring of multiple myeloma in conjunction with other laboratory and clinical findings. For *in-vitro* diagnostic use only.

VI Standards/Guidance Documents Referenced:

Not applicable

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

   Refer to K153394

2. Linearity:

   Refer to K211648

3. Analytical Specificity/Interference:

   Refer to K153394

4. Assay Reportable Range:

   See K153394, K211648

5. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

   Refer to K153394

6. Detection Limit:

   Refer to K211648

7. Assay Cut-Off:

   Refer to K153394 and K184338

B Comparison Studies:

1. Method Comparison with Predicate Device:

   Not applicable

2. Matrix Comparison:

   Not applicable

C Clinical Studies:

Clinical performance of the Diazyme Human Kappa (κ) Free Light Chain Assay and the Diazyme Human Lambda (λ) Free Light Chain Assay as an aid in the diagnosis and monitoring of multiple myeloma was previously demonstrated (Refer to K153394 and K220001).

The performance of the Diazyme Human Kappa (κ) Free Light Chain Assay and the Diazyme Lambda (λ) Free Light Chain Assay as an aid in the evaluation of MGUS was investigated from the following studies:

Study 1:

A study was performed by testing a total of 270 serum samples from clinically confirmed MGUS patients (with IFE positivity) and a total of 266 samples from clinically defined non-MGUS patients with polyclonal immunostimulation (confirmed with negative SPEP/SIFE). Clinical diagnostic criteria and classification for MGUS and related plasma-cell disorders were, as practiced clinically, fulfilled, but were not limited to the criteria outlined by the 'International Myeloma Working Group (IMWG)' consensus. The result of the device was compared to the clinical diagnosis for each sample.

The cohort of 270 MGUS samples included 205 non-IgM MGUS, 40 IgM MGUS and 25 light chain (LC) MGUS. All samples were tested for FLC kappa and lambda levels with the candidate devices, the Diazyme Human Kappa (κ) Free Light Chain Assay and the Diazyme Human Lambda (λ) Free Light Chain Assay on the Roche cobas c 501 analyzer. FLC κ/λ ratios were calculated for each sample. The test results for MGUS positive or negative were based on the following criteria:

- For Non-LC MGUS, the test was considered as "MGUS Positive" or "Abnormal" when abnormal FLC κ/λ ratio (outside reference interval, i.e., < 0.22 or > 1.74) with IFE positivity was determined.

- For LC MGUS, the test was considered as "MGUS Positive" when abnormal FLC κ/λ ratio (outside reference interval, i.e., < 0.22 or > 1.74) with elevated FLC kappa (>20.7 mg/L) or elevated FLC lambda FLC (> 27.7 mg/L) was determined.

Results:

The results showed that 145 out of 270 MGUS cases were tested positive by the Diazyme Human Kappa (κ) Free Light Chain Assay and the Diazyme Human Lambda (λ) Free Light Chain Assay with a positive rate of 53.7% (95% CI: 47.6% – 59.8%). The distribution of the cohort and the positivity rate for each clinical type of MGUS are summarized in the following table:

MGUS Type	N	n (n/N%) of MGUS Positive by Diazyme FLC Assays
Non-IgM MGUS (all)	**205**	**92 (44.9%)**
IgG κ	85	48 (56.5%)
IgG λ	80	26 (32.5%)
IgA κ	25	13 (52.0%)
IgA λ	15	5 (33.3%)
IgD κ	0	0 (0%)
IgD λ	0	0 (0%)
IgM MGUS (all)	**40**	**28 (70.0%)**
IgM κ	30	24 (80.0%)
IgM λ	10	4 (40.0%)
LC-MGUS (all)	**25**	**25 (100%)**
κ	15	15 (100%)
λ	10	10 (100%)
TOTAL	**270**	**145 (53.7%)**

The 266 samples from patients with confirmed non-MGUS (negative SPE/IFE) polyclonal stimulation (polyclonal hypergammaglobulinemia) were comprised of: connective tissue disease (59); renal disease (46); infection (40); CVD (21); neurological diseases (20); dermatological disease (15); cancers (14); autoimmune diseases (13); B Lymphoma (8);

anemia (6); liver disease (5); diabetes (4) and other miscellaneous diseases/conditions (15) including: polyarteritis nodosa, vascular disease, gout, immunodeficiency, venous insufficiency, thrombocytosis, leukocytosis, thrombotic microangiopathy, lymphadenopathy, COPD, pulmonary fibrosis, interstitial lung disease, lung nodules.

Among 266 non-MGUS samples, 250 of these were determined as negative by the Diazyme Kappa and Lambda FLC assays, indicating a negative agreement of 94.0% (95% CI: 90.4%-96.5%) in this sample cohort. The 16 false positive samples were from patients with the following disorders: cancers (4); renal diseases (3); connective (2); anemia (2); infection/inflammation (2); autoimmune diseases (1); B lymphoma (1); and other diseases/conditions (1).

Study 2:

Another study was performed by testing 348 serum samples from 84 subjects with clinically stable MGUS and two subjects with progressive clinical status converting from MGUS to multiple myeloma (MM). Among 84 MGUS stable subjects, 69 patients were diagnosed with non-IgM MGUS (29 IgG K, 22 IgG L, 11 IgA K, and seven IgA L), and 11 patients with IgM MGUS (eight IgM K and three IgM L), and four patients with LC MGUS (one LC-K, three LC-L). The two progressive subjects were one IgG L and one LC-L.

At least three and up to five serial draws were collected from each patient and tested with the Diazyme Human Kappa (κ) Free Light Chain Assay and the Diazyme Human Lambda (λ) Free Light Chain Assay on the Roche cobas c 501 analyzer. FLC κ/λ ratio was calculated for each blood draw sample. Since the MGUS has been identified as a precursor to malignant diseases (multiple myeloma or amyloidosis), but not as a disease which requires treatment, no defined criteria in how to interpret consecutive FLC results are available. Therefore, for device evaluation purposes only, the criteria for stable MGUS and progressive MGUS based on test results in this study are defined as follows:

- Stable MGUS defined as < 25% increase in the concentration of the involved FLC in two consecutive assessments. This analysis included MGUS patients with and without abnormal FLC κ/λ ratio.

- Progressive MGUS defined as the FLC κ/λ ratio outside of the reference interval of 0.22 – 1.74, and an increase of ≥ 25% in the concentration of the involved light chain at or preceding the diagnosis of MM, for two consecutive assessments.

Results:

Seventy-six (76) out of 84 clinically stable MGUS subjects were determined as stable by the test and two out of two clinically progressive subjects were determined as progressive by these tests.

Limitations

- The performance has not been fully evaluated on all race/ethnicity in the intended use population.
- The study included time points of blood draws not indicative of clinical practice. The algorithm using only FLC has not been validated for progression of disease. Furthermore, a small sample size (two patients) with the subtypes Lambda IgG and LC-L was used to study

progression in Study 2. Risk mitigation strategies include that this test is not a stand-alone test for the evaluation of patients with MGUS and the test is to be utilized in conjunction with serum protein electrophoresis and immunofixation blood tests.

D Clinical Cut-Off:

Not applicable

E Expected Values/Reference Range:

Refer to K153394 and K181438

Kappa FLC reference range: 2.4 – 20.7 mg/L
Lambda FLC reference range: 4.2 – 27.7 mg/L
Kappa/Lambda Ratio reference Range: 0.22 – 1.74

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.