510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY

A. 510(k) Number:
k083846

B. Purpose for Submission:
New Device

C. Measurand:
CFTR (Cystic Fibrosis transmembrane conductance regulator) gene from human blood specimens

D. Type of Test:
Multiplex PCR followed by multiplex allele specific primer extension for genotyping, hybridized to multiplexed fluorescing microparticles, detected by flow cytometry

E. Applicant:
Luminex Molecular Diagnostics Inc.

F. Proprietary and Established Names:
xTAG™ Cystic Fibrosis 39 Kit v2

G. Regulatory Information:

1. Regulation section:
   21 CFR 866.5900, CFTR (cystic fibrosis transmembrane conductance regulator) gene mutation detection system

2. Classification:
   Class II

3. Product code:
   NUA, system, test, CFTR (cystic fibrosis transmembrane conductance regulator) gene mutation detection

4. Panel:
   Immunology (82)

H. Intended Use:

1. Intended use(s):
   The xTAG™ Cystic Fibrosis 39 Kit v2 is a device used to simultaneously detect and identify a panel of mutations and variants in the cystic fibrosis transmembrane conductance regulator (CFTR) gene in human blood specimens. The panel includes mutations and variants currently recommended by the American College of Medical Genetics and American College of Obstetricians and Gynecologists (ACMG/ACOG) plus some of the world's most common and North American prevalent mutations. The xTAG™ Cystic Fibrosis 39 Kit v2 is a qualitative genotyping test which provides information intended to be used for carrier testing in adults of reproductive age, as an aid in newborn screening, and in confirmatory diagnostic testing in newborns and children.

   The kit is not indicated for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes.

2. Indication(s) for use:
   Same as Intended use.

3. Special conditions for use statement(s):
   For Prescription use only

The kit is not indicated for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes

4. Special instrument requirements:
Luminex 100 or 200 IS instruments with IS v2.3 (see k073506 for details) and xPONENT v3.1

I. Device Description:

The xTAG™ Cystic Fibrosis 39 Kit v2 is comprised of the following components: 1 vial (240 µL) xTAG CFTR PCR Primer Mix v2; 1 vial (192 µL) xTAG CFTR ASPE Primer Mix A v2; 2 vials (5 units/µL,115 µL each) Platinum Tfi Exo(-) DNA Polymerase; 4 vials (1.3 mL each) 5X Platinum Tfi Reaction buffer; 2 vials (1 mL each) Tfi 50 mM $MgCl_2$; 2 vials (10 units/µL, 48 µL each) xTAG Exonuclease I; 2 vials (1 unit/µL, 120 µL each) xTAG Shrimp Alkaline Phosphatase (SAP); 1 vial (1 mg/mL, 108 µL) xTAG Streptavidin, R-Phycoerythrin Conjugate; 1 vial (2.16 mL) xTAG CFTR Bead Mix A v2; and 1 bottle (12 mL) xTAG 10X Buffer.

J. Substantial Equivalence Information:

1. Predicate device name(s):
xTAG™ Cystic Fibrosis Kit

2. Predicate 510(k) number(s):
k043011, k060627

3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Intended Use	The xTAG™ Cystic Fibrosis 39 Kit v2 is a device used to simultaneously detect and identify a panel of mutations and variants in the cystic fibrosis transmembrance conductance regulator (CFTR) gene in human blood specimens. The panel includes mutations and variants currently recommended by the American College of Medical Genetics and American College of Obstetricians and Gynecologists (ACMG/ACOG), plus some of the world's most common and North American prevalent mutations.	Same
Indications for Use	The xTAG™ Cystic Fibrosis 39 Kit v2 is a qualitative genotyping test which provides information intended to be used for carrier testing in adults of reproductive age, as an aid in newborn screening, and in confirmatory diagnostic testing in newborns and children.	Same
Contra-Indications	This kit is not intended for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes.	Same
Type of Test	Multiplex PCR followed by multiplex allele specific primer extension for genotyping, hybridized to multiplex fluorescent microparticles, detected by flow cytometry	Same
Product Description	Tests for 39 mutations and 4 variants in the CFTR gene (23 of which are recommended by ACMG/ACOG).	Same
Specimen type	Peripheral human whole blood	Same

Differences		
Item	Device	Predicate
Software	TDAS CFTR contains 1 template to detect a panel of 39 mutations and 4 variants. Software has a masking function where user can choose to display results for either the ACMG/ACOG 23 mutations or for the full panel of mutations.	TDAS CF-1 contains 1 template to detect a panel of 39 mutations and 4 variants only.
Instrument system	Luminex 100 or 200 IS v.2.3	Luminex 100 IS v2.3
Instrument software	IS v2.3 and xPONENT v3.1	IS v2.3 only

K. Standard/Guidance Document Referenced (if applicable):
- American College of Medical Genetics (ACMG) / American College of Obstetricians and Gynecologists Technical Standards and Guidelines for CFTR Mutation Testing and Standards and Guidelines for Clinical Genetic Laboratories
- Cystic Fibrosis Foundation / Center for Disease Control Recommendations on Newborn Screening for CF
- FDA Class II Special Controls Guidance: Quality Control Material for Cystic Fibrosis Nucleic Acid Assays (Jan 2007)
- FDA Class II Special Controls Guidance: CFTR Gene Mutation Detection Systems (Oct 2005)
- CDRH Draft Guidance on Multiplex Tests for Heritable DNA Markers, Mutations and Expression Patterns (Feb 2003)
- CDRH Draft Guidance on Statistical Guidance on Reporting Results from Studies Evaluating Diagnostic Tests (Mar 2003)
- CDRH Guidance for the Content of Pre-Market Submission for Software Contained in Medical Devices (May 1998)
- CDRH Guidance on General Principles of Software Validation (Jan 2002)
- CDRH Guidance on Format for Traditional and Abbreviated 510ks (Aug 2005)
- MM01-A2: Molecular Diagnostic Methods for Genetic Diseases
- MM13-PE: Collection, Transport, Preparation, and Storage of Specimens for Molecular Methods
- MM17-A: Verification and Validation of Multiplex Nucleic Acid Assays
- EP05-A2: Evaluation of Precision Performance of Clinical Chemistry Devices
- EP07-A2E: Interference Testing in Clinical Chemistry
- EP12-A: User Protocol for Evaluation of Qualitative Test Performance
- EP17-A: Protocols for Determining Limits of Detection and Limits of Quantitation

L. Test Principle:
The xTAG™ CFTR 39 Kit v2 incorporates multiplex Polymerase Chain Reaction (PCR) and multiplex Allele Specific Primer Extension (ASPE) with Luminex's proprietary Universal Tag sorting system on the Luminex® 100 or 200 xMAP™ platform.

The amplimer sizes range from 179 bp to 465 bp. Purified genomic DNA is extracted from each sample and a multiplex PCR reaction is carried out under optimized conditions. Each sample undergoes a multiplex allele specific primer extension (ASPE) reaction, where an aliquot of the PCR product is run through the ASPE A reaction. This step allows for detection of each allele (wild-type or mutant) of a given locus using an allele-specific probe (ASP) which contains a unique DNA sequence (tag) at its 5' end. Each bi-allelic locus has 2 ASPs and each tri-allelic loci has 3 ASPs included in the ASPE Mix. For each ASP, the 3' end of the primer is a perfect match for its allele, but will have a 3' mismatch on any other allele. Both these ASPs however are tagged with a common tag at their 5' end. The DNA polymerase will only extend the primer when there is a perfect match on the 3' end, so that the primer is only extended if its target allele is present in the sample. Biotin-dCTP is incorporated into the extending chain if extension occurs.

For the hybridization reaction, the ASPE reaction product is added directly to microwells containing aliquots of the Bead Mix. Each coupled bead is spectrally distinguishable from the other coupled beads in a given bead mix. A fluorescent reporter molecule (streptavidin-phycoerythrin) is bound to the biotin on the extended primers. Each tagged primer hybridizes only to its unique anti-tag complement; therefore, each colored bead represents a specific allele, through the bead/anti-tag/tagged primer association. The beads are then analyzed by the Luminex instrument, which contains two lasers: one identifies the color-coded bead, and the other identifies the presence or absence of extended allele specific primer through the phycoerythrin reporter. Thus, the genotype of that locus is identified by the presence of phycoerythrin signal attached to one or both ASPs.

For each sample analyzed by the CFTR 39 kit v2, an output file containing MFI signals from the Luminex Instrument is generated. The proprietary software component of this product analyzes this output data file, to provide a final qualitative genotype for the sample. The software includes a feature which enables users to select between 2 options for the final output:

Option 1: Full Panel (39 mutations + 4 variants).
Option 2: ACMG/ACOG panel (23 mutations).

When Option 2 is chosen, the non-ACMG/ACOG recommended panel of mutations/variants will not be available to the user.

Mutations and 4 variants (variants italized) included in the CFTR 39 Kit v2

ΔF508*	1717-1G>A*	W1282X*	2307insA
*ΔI507**	R560T*	1078delT	Y1092X
G542X*	R553X*	394delTT	M1101K
G85E*	G551D*	Y122X	S1255X
R117H*	1898+1G>A*	R347H	3876delA
621+1G>T*	2184delA*	V520F	3905insT
711+1G>T*	2789+5G>A*	A559T	*5/7/9T*
N1303K*	3120+1G>A*	S549N	*F508C*
R334W*	R1162X*	S549R	*I507V*
R347P*	3659delC*	1898+5G>T	*I506V*
A455E*	3849+10kbC>T*	2183AA>G	

M. Performance Characteristics (if/when applicable):
1. <u>Analytical performance:</u>
   a. *Precision/Reproducibility:*
   A multi-centre, multi-operator, multi-lot, blinded study design was used to evaluate total variability of the xTAG CFTR v2 system. The experimental design generally follows CLSI recommendations (EP5-A and MM17-A) and requirements laid forth by the FDA in I080024.

   The study included two arms. **Arm A** evaluated the reproducibility of the sample extraction step using 18 unique clinical (whole blood) samples representing wild-type and 3 mutant genotypes. DNA was extracted using 3 different methods and tested using the same assay lot by 2 operators, across 9 non-consecutive days. Each operator performed 3 runs/extraction methods, and each assay point was run in duplicate. An extraction was performed for each assay run by each operator. The number of replicates per sample was: (3 extraction methods) x (2 operators/extraction method) x (3 runs/operator) x (2 replicates/run) = 36 replicates. The results are summarized in the following table.

   Summary of Results from Reproducibility Study Arm A

Extraction Method	Number of Samples	Total Number of Assays	CFTR v2 Results
a	18	18 x 12 = 216	All assays passed (correct calls)
b	18	18 x 12 = 216	All assays passed (correct calls)
c	18	18 x 12 = 216	All assays passed (correct calls)

   Arm B evaluated the reproducibility of the analytical (post-extraction) steps of the assays at 3 external sites using in order of preference and availability, purified genomic DNAs extracted from clinical (whole blood) samples, purified genomic DNA extracted from lymphoid cell lines, and/or plasmids. Each set contained samples representing all mutations and variants probed by the CFTR 39 Kit v2. There were 2 operators per site, each performing 1 run/day across 3 non-consecutive days (3 runs per operator or 6 runs per site). Within a given run, each assay point was run in duplicate. A total of three assay lots were tested (1 lot/site). This arm of the study also provided information on the ability to resolve different sources of variability in the post-extraction (analytical) step of the assay. For example, it was possible to analyze data from a particular site to estimate the operator-to-operator variability.

   The results indicated a precision of >99.54% across the 3 sites. There was one "no-call" after 1 re-run at Site 3, and 3 mis-calls (from a single plasmid sample) at Site 1. Reproducibility of a compound heterozygote dF508/F508C was also demonstrated in

this study. 36 of 30 replicates of one sample generated a dF508 call and 6 generated a dF508 Mu D call (Mu D calls are defined as detection of only a mutant allele).

Between Site/ Operator Reproducibility Data for xTAG CFTR 39 Kit v2

Sample ID	Genotype		Operator - to – Operator											
			Site 1				Site 2				Site 3			
			†Op 1 N ¥	Op 1 % corr ‡	Op2 N	Op2 % corr	Op1 N	Op1 % corr	Op2 N	Op2 % corr	Op1 N	Op1 % corr	Op2 N	Op2 % corr
3893	711+1G>T	dF508	438	100	438	100	438	100	438	100	438	100	438	100
4919	1717-1G>A	-	438	100	438	100	438	100	438	100	438	100	438	100
9354	G542X	R117H	438	100	438	100	438	100	438	100	438	100	438	100
10561	A455E	-	438	100	438	100	438	100	438	100	438	100	438	100
10839	3659delC	-	438	100	438	100	438	100	438	100	438	100	438	100
10988	R1162X	dF508	438	100	438	100	438	100	438	100	438	100	438	100
11688	3849+10kbC>T	-	438	100	438	100	438	100	438	100	438	100	438	100
15249	W1282X	-	438	100	438	100	438	100	438	100	438	100	438	100
15560	1078delT	dF508	438	100	438	100	438	100	438	100	438	100	438	100
17594	A559T	-	438	100	438	100	438	100	438	100	438	100	438	100
30116	S549N	-	438	100	438	100	438	100	438	100	438	100	438	100
41406	G551D	R347P	438	100	438	100	438	100	438	100	438	100	438	100
48308	3905insT	-	438	100	438	100	438	100	438	100	438	100	438	100
53948	R560T	dF508	438	100	438	100	438	100	438	100	438	100	438	100
63436	394delTT	-	438	100	438	100	438	100	438	100	438	100	438	100
69711	R553X	-	438	100	438	100	438	100	438	100	438	100	438	100
70268	2184delA	-	438	100	438	100	438	100	438	100	438	100	438	100
71713	1898+1G>A	dF508	438	100	438	100	438	100	438	100	438	100	438	100
77590	Y1092X-C>A	dF508	438	100	438	100	438	100	438	100	438	100	438	100
89633	2183AA>G	-	438	100	438	100	438	100	438	100	438	100	438	100
98748	V520F	3120+1G>A	438	100	438	100	438	100	438	100	438	100	438	100
LMD002-08 111	R334W	-	438	100	438	100	438	100	438	100	438	100	438	100
LMD002-08 33	2789+5G>A	-	438	100	438	100	438	100	438	100	438	100	438	100
LMD002-08 58	612+1 G>A	-	438	100	438	100	438	100	438	100	438	100	438	100
LMD002-08 64	dI507	-	438	100	438	100	438	100	438	100	438	100	438	100
LMD002-08 73	dF508 (+ F508C variant)	-	439*	100	442**	100	438***	100	438***	100	438***	100	439*	100.00
LMD002-08 86	G85E	-	438	100	438	100	438	100	438	100	438	100	438	100
LMD002-08 90	N1303K	-	438	100	438	100	438	100	438	100	438	100	438	100
NA07857	M1101K	M1101K	438	100	438	100	438	100	438	100	438	100	438	100
NA18802	Y122X	R1158X	438	100	438	100	438	100	438	100	438	100	438	100
NA20737	R347H	-	438	100	438	100	438	100	438	100	438	100	438	100
NA20741	3876delA	-	438	100	438	100	438	100	438	100	438	100	438	100

			N	% corr	N	% corr	N	% corr	N	% corr	N	% corr	N	% corr
NA20915	S549R	-	438	100	438	100	438	100	438	100	438	100	438	100
NA21551	dF508	-	438	100	438	100	438	100	438	100	438	100	437	99.77
Plasmids 10B+10C‡‡	dF508(+I506V variant)	V520F	18	100	18	100	18	100	18	100	18	100	18	100
Plasmid 12B	1898+5G>T	-	6	100	6	100	6	100	6	100	6	100	6	100
Plasmid 13E	2307insA	2055del9>A	12	91.67	12	83.33	12	100	12	100	12	100	12	100
Plasmid 19E	3791delC	-	12	100	12	100	12	100	12	100	12	100	12	100
Plasmid 17bB	Y1092X-C>G	-	6	100	6	100	6	100	6	100	6	100	6	100
Plasmid 19A	S1255X (ex.19)	-	6	100	6	100	6	100	6	100	6	100	6	100
Plasmid 20A	S1255X (ex.20)	W1282X	12	100	12	100	12	100	12	100	12	100	12	100

Site 1: Hartford Hospital, Connecticut, USA; Site 2: Luminex Molecular Diagnostics, Toronto, CA; Site 3: Hospital for Sick Children, Toronto, CA

† Op = operator (1 or 2)

¥ N = number of calls

‡ % corr = percent correct

* Total number of calls 438 + 1 = 439, because TDAS made one dF508 Mu D call (and the F508C variant call is unmasked)

** Total number of calls 438 + 4 = 442, because TDAS made 4 dF508 Mu D calls (and the F508C variant call is unmasked)

*** Total number of calls = 438, because TDAS made all dF508 HET calls (the F508C variant call is masked)

b. *Linearity/assay reportable range:*
Not Applicable

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*
Previous stability studies support a shelf-life of 1 year when kit reagents are stored at -25°C to -15°C. Repeated freeze-thaw cycles (up to 4) do not compromise the integrity of the xTAG™ CFTR 39 Kit v2.
Assay controls: The package insert recommends that at least two negative controls and a rotating set of positive controls (including a dF508 mutation) are included with each run.

d. *Detection limit:*
The package insert recommends that purified DNA have at least a UV 260/280 ratio of >1.5 depending on the extraction method used. The assay has been optimized for use with 50 ng of total DNA, but reliable results were previously demonstrated with DNA amounts between 10 ng to 1.5 µg.

The detection limits were determined by serially diluting 10 gDNA samples, obtained from commercially available cell lines at 10 levels ranging from 300 - 0.39 ng/µL. The LoD is defined as the lowest amount of genomic DNA in a sample for which the assay can detect genotypes with a positive agreement rate of ≥ 95%. Eight of the samples were heterozygous, one (1) was homozygous, and one (1) contained a non-panel mutation which would be called as wild type by the assay. Three of the heterozygous samples were compound heterozygotes, but the second allele was a non-panel mutation and therefore called as wild-type by the assay. At each

concentration, 22 replicates from each of the samples were run. Based on initial results, a LoD = 1.56 ng/µL was proposed. An additional study was performed using 2 ng/µL for each of the 22 sample replicates. Two samples exhibited 21/22 correct calls, corresponding to a 95.4% positive agreement. No failures were observed at the 300 ng/µL concentration.

    e. *Analytical specificity:*

To examine the effects of potential interferents that might be found in whole blood samples, a study was conducted using 8 whole blood samples (4 wild-type for CFTR mutations, 1 N1303K Het, 1 V520F Het, and 2 ΔF508 Het). Each sample was split into 6 parts, and incubated in the absence or presence of one of 3 potential interferents (hemoglobin at final concentration of 1,500 µg/mL, bilirubin at a final concentration of 200 µg/mL, and a mixture of triglycerides at a final concentration of 30 mg/mL). Samples were extracted and assayed with the kit according to the instructions. Dideoxy- sequencing of the CFTR exons in each sample was conducted to confirm the genotypes of each sample. Results indicated that none of the substances at the concentrations tested produced a significant inhibitory effect on kit performance.

    f. *Assay cut-off:*

Not Applicable

2. Comparison studies:

    a. *Method comparison with predicate device:*

Accuracy of the xTAG CFTR 39 Kit v2 was assessed through evaluation of samples representing all alleles (mutations and polymorphisms) probed by the assay. Three hundred nineteen (319) samples were used in this method comparison study. These samples were left-over, anonymized, clinical specimens, eight were genomic DNAs from EBV-transformed lymphoid cell lines, and 2 were custom-designed plasmids engineered to contain 1-2 CFTR mutations. The genotype of all samples was confirmed by bi-directional sequencing. The xTAG Cystic Fibrosis Mutation Detection Kit was used as the comparator method.

For all exons, the results indicated 100% agreement between the xTAG Cystic Fibrosis 39 Kit v2 and the xTAG Cystic Fibrosis Mutation Detection Kit. There were 0 no-calls and 0 mis-calls in the accuracy study.

Table: Overall accuracy for the xTAG Cystic Fibrosis 39 Kit v2

Exon or Intron	Mutations	Number of Samples tested per mutation			Before Allowable Rerun			After Allowable Rerun				
		Independent Clinical Samples	Cell lines	Plasmids	Repeats due to mis-calls	Repeats due to no-calls	% Accuracy prior to repeats	Repeats due to mis-calls	Repeats due to no-calls	Final % Accuracy (after repeats)	LB of 95% CI*	UB of 95% CI*
Ex 3	G85E#	2	0	0	0	0	100	0	0	100	15.81	100
	394delTT	2	0	0	0	0	100	0	0	100	15.81	100
Ex 4	R117H	36	0	0	0	0	100	0	0	100	90.51	100
	Y122X	1	1	0	0	0	100	0	0	100	15.81	100
	621+1G>T#	6	0	0	0	0	100	0	0	100	54.07	100
Ex 5	711+1G>T#	3	0	0	0	0	100	0	0	100	29.24	100
Ex 7	1078delT	3	0	0	0	0	100	0	0	100	29.24	100
	R334W#	3	0	0	0	0	100	0	0	100	29.24	100
	R347Pmut#	6	0	0	0	0	100	0	0	100	54.07	100

		Number of Samples tested per mutation			Before Allowable Rerun			After Allowable Rerun				
	R347Hmut	3	1	0	0	0	100	0	0	100	39.76	100
Ex 9	A455E#	3	0	0	0	0	100	0	0	100	29.24	100
Ex 10	Dl507mt#	9	0	0	0	0	100	0	0	100	66.37	100
	dF508mut#	162	1	0	0	0	100	0	0	100	97.87	100
	V520F	2	0	0	0	0	100	0	0	100	15.81	100
Ex 11	1717-1G>A#	5	0	0	0	0	100	0	0	100	47.82	100
	G542X#	13	0	0	0	0	100	0	0	100	75.29	100
	S549N	1	1	0	0	0	100	0	0	100	15.81	100
	S549R	2	1	0	0	0	100	0	0	100	47.82	100
	G551D#	12	0	0	0	0	100	0	0	100	73.54	100
	R553X	7	0	0	0	0	100	0	0	100	59.04	100
	A5559T	2	0	0	0	0	100	0	0	100	29.24	100
	R560T#	4	0	0	0	0	100	0	0	100	36.76	100
Ex12	1898+1G>A#	2	0	0	0	0	100	0	0	100	15.81	100
	1898+5G>T	0	0	2	0	0	100	0	0	100	15.81	100
Ex13	2183AA>G	2	0	0	0	0	100	0	0	100	15.81	100
	2184delA#	1	0	0	0	0	100	0	0	100	2.50	100
	2307insA	3	0	0	0	0	100	0	0	100	29.24	100
Ex 14b	2789+5G>A#	5	0	0	0	0	100	0	0	100	47.82	100
Ex 16	3120+1G>A	7	0	0	0	0	100	0	0	100	59.04	100
Ex17b	Y1092X-C>G	0	0	2	0	0	100	0	0	100	15.81	100
	Y1092X-C>A	2	0	0	0	0	100	0	0	100	15.81	100
	M1101K	0	2	0	0	0	100	0	0	100	15.81	100
Ex 19	R1162X#	5	0	0	0	0	100	0	0	100	47.82	100
	3659delC#	4	0	0	0	0	100	0	0	100	39.76	100
	S1255X(19)	4	0	0	0	0	100	0	0	100	39.76	100
Intron 19	3849+10kb#	13	0	0	0	0	100	0	0	100	75.29	100
Ex 20	S1255X(20)	4	0	0	0	0	100	0	0	100	39.76	100
	3876delA	1	1	0	0	0	100	0	0	100	29.24	100
	3905insT	2	0	0	0	0	100	0	0	100	15.81	100
	W1282X#	8	0	0	0	0	100	0	0	100	63.06	100
Ex 21	N1303K#	6	0	0	0	0	100	0	0	100	54.07	100
Ex 10	I506V-var tg	3	0	0	0	0	100	0	0	100	29.24	100
Ex 10	I506V-variant	5	0	0	0	0	100	0	0	100	47.82	100
	I507V-variant	0	1	0	0	0	100	0	0	100	2.50	100
	F508C-variant	5	0	0	0	0	100	0	0	100	47.82	100

\# member of ACMG-23 panel

*Clopper-Pearson CI calculator provided by John C. Pezzullo (Kissimmee, Florida, USA) and is available at http://statpages.org/confint.html

    b. *Matrix comparison:*
      Not applicable. This test is only for use with human whole blood collected using EDTA as the anticoagulant.

3. Clinical studies:
    a. *Clinical Sensitivity:*

The clinical sensitivity can be estimated based on the published studies of mutation frequencies in various ethnicities and based on the results of analytical studies described in this submission.

b. *Clinical specificity*:
The clinical specificity can be estimated based on the published literature and the results of analytical studies described in this submission.

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:
Cystic fibrosis (CF) is the most common autosomal recessive disorder in the Caucasian population, with an incidence of approximately 1 in 3,200 live births. The incidence of CF in other ethnic groups varies, as seen in the following table.

Ethnic Group	Incidence of Cystic Fibrosis
North American Caucasian	1 in 3200
Ashkenazi Jewish	1 in 3300
Hispanic	1 in 9500
African American	1 in 15 300
Asian American	1 in 32 100
Native American (Pueblo)	1 in 3970
Native American (Zuni)	1 in 1347

N. Instrument Name:
Luminex System (100 IS or 200 including ISv2.3 or xPONENT v3.1 software, calibrators, and controls)

O. System Descriptions:
See k073506 for a description of the Luminex System and base software environment

Results:
For each sample analyzed by the CFTR 39 kit v2, an output file containing MFI signals from the Luminex Instrument is generated. The proprietary software component of this product analyzes this output data file, to provide a final qualitative genotype for the sample. The software includes a feature which enables users to select between 2 options for the final output:

Option 1: Full Panel (39 mutations/deletions + 4 variants).
Option 2: ACMG/ACOG panel (23 mutations and deletions).

When Option 2 is chosen, the non-ACMG/ACOG recommended panel of mutations/variants will not be available to the user.

FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

Yes ____x____ or No _____
1. Specimen Identification:
    Users must fill in Batch Information by providing a unique batch Name, Description and Creator. Users have to enter appropriate patient information, i.e. number of samples, and sample IDs.
2. Specimen Sampling and Handling:
    Genomic DNA preparation, multiplex (16-plex) PCR, amplicon treatment, multiplex ASPE, bead hybridization and incubation with reporter are performed semi-manually. Subsequently, beads are analyzed by the Luminex® 100 xMAP™ system, and data generated analyzed by TDAS CFTR Analysis Software CF-I (TDAS CF-I) to provide final genotype for the sample. User must pre-select sample view, either "ACMG panel" or "Full Panel" for each unique patient sample to be analyzed. The user will not be able to unmask any hidden data for samples for which the "ACMG panel" is selected, and visa-versa.
3. Calibration:
    The Luminex system should be prepared and calibrated according to procedures described in the Luminex 100 IS 2.3 user Manual or Luminex 200 System user Manual.

    The xTAG Cystic Fibrosis v2 T-A IS template must be installed on the computer that controls the Luminex system.
4. Quality Control:
    Negative Controls:
    At least two DNase, RNase free water controls are required with each run. The wells which are negative controls must be designated when analyzing data with the TDAS CFTR software.

    Positive Controls:
    It is recommended to routinely include in each run a set of rotating positive controls for CFTR mutations probed by the kit. Since the CF dF508 mutation is the most common (accounting for 30-88% of all CF mutations depending on ethnic group*) it is recommended that a control sample wit this mutation be included in every run. Luminex Molecular Diagnostics recommends the use of genomic DNA controls similar to the specimen type whenever feasible although spiked controls (using synthetic DNA) may be used when specimen samples are not available.
    (*Gibson, Moskowitz et al, 2001)

P. Other Supportive Instrument Performance Characteristics Data Not Covered In The "Performance Characteristics" Section above:
None

Q. Proposed Labeling:
The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10, labeling for in vitro diagnostic products.

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