510(K) SUMMARY

AUG 2 6 2010

xTAG® CYP2D6 Kit v3

This summary of 510(k) safety and effectiveness information is being submitted in accordance with the requirement of 21 CFR 807.92.

510(k) Number:
K093420

Purpose for Submission:
New Device.

Measurand:
A panel of nucleotide variants found within the highly polymorphic CYP2D6 gene located on chromosome 22 from genomic DNA extracted from whole blood sample. This kit can also identify gene rearrangements associated with the deletion (*5) and duplication genotypes.

Type of Test:
Qualitative genotyping multiplex test.

Applicant:
Luminex Molecular Diagnostics Inc.
439 University Ave.
Toronto, ON M5G 1Y8 Canada
Tel: 416.593.4323 x374
Fax: 416.593.1001
Contact person: Dr. Gloria Lee

Proprietary and Established Names:
xTAG® CYP2D6 Kit v3

Regulatory Information:
1. Regulation Section:
21 CFR 862.3360 - Drug Metabolizing Enzyme Genotyping System

2. Classification:
Class II

3. Product Code:
NTI

4. Panel:
Toxicology

Intended Use:
The xTAG® CYP2D6 Kit v3 is a device used to simultaneously detect and identify a panel of nucleotide variants found within the highly polymorphic CYP2D6 gene located on chromosome 22 from genomic DNA extracted from EDTA and citrate anticoagulated whole blood samples. This kit can also identify gene rearrangements associated with the deletion (*5) and duplication genotypes. The xTAG® CYP2D6 Kit v3 is a qualitative genotyping assay which can be used as an aid to clinicians in determining therapeutic strategy for therapeutics that are metabolized by the CYP2D6 gene product. This kit is not indicated for stand-alone diagnostic purposes. This test is not intended to be used to predict drug response or non-response.

Indication(s) for use: Same as intended use.

Special conditions for use statement(s):
This kit is not indicated for stand-alone diagnostic purposes. This test is not intended to be used to predict drug response or non-response.

Special instrument requirements:
Luminex® 100/200 Instrument

Device Description:
The xTAG CYP2D6 Kit v3 includes the following components:

- xTAG® 2D6 v3 PCR Primer Mix A
- xTAG® 2D6 v3 PCR Primer Mix B
- xTAG® 2D6 v3 ASPE Primer Mix
- xTAG® 2D6 v3 Bead Mix
- xTAG® 10x Buffer
- xTAG® Shrimp Alkaline Phosphatase
- xTAG® Exonuclease I
- xTAG® Streptavidin, R-Phycoerythrin Conjugate
- Platinum® Tfl Exo(-) DNA Polymerase
- Platinum® Tfl Reaction Buffer, 5x
- Tfl 50mM $MgCl_2$
- xTAG® Hot Start Taq (Long Acting)
- xTAG® 10x Taq Buffer (Long Acting)
- xTAG® Data Analysis Software (TDAS) CYP2D6

Substantial Equivalence Information:

1. Predicate device name(s):
AmpliChip CYP450 Test.

2. Predicate 510(k) number(s):
K042259

Standard/Guidance Document Referenced (if applicable):

- 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 on Pharmacogenetic Tests and Genetic Tests for Heritable Markers (June, 2007)
- 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
- 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

Test Principle:

The CYP2D6 assay 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 / 200 Instrument. Raw data (mean fluorescence intensity, MFI, signals) are analyzed by the xTAG® Data Analysis Software (TDAS) and result outputs are provided as qualitative calls.

For each genomic sample being tested, two separate PCR reactions are performed. PCR-A produces an alpha fragment (3.8 kb) used to detect the mutations in Table 1, as well as a duplication amplimer (3.2 kb) which indicates the presence of the duplication genotype. PCR-B produces a beta fragment (2.6 kb) used to detect the mutations in Table 1, as well as a deletion amplimer (3.5 kb) indicative of the deletion genotype. Following PCR amplification, the two reactions (PCR-A and PCR-B) are pooled.

To enable efficient incorporation of biotin-dCTP during the ASPE reaction, the pooled PCR product is treated with Shrimp Alkaline Phosphatase to inactivate any remaining nucleotides (particularly dCTP), and with Exonuclease I to degrade any primers left over from the PCR reaction.

ASPE is then carried out using universally-tagged primers supplied in the ASPE primer mix. An aliquot of the ASPE reaction is hybridized with the universal array (Bead Mix) in the presence of the hybridization buffer and incubated with Streptavidin, R-Phycoerythrin conjugate (reporter solution).

Samples are read on the Luminex® 100/200 Instrument and signals are generated for each of the loci as well as for the duplication and deletion amplimers, if present. These fluorescence values are then analyzed to determine whether the wild-type/mutant allele for each of the loci has been detected, or whether the samples carry an allele(s) with the deletion or duplication.

Table 1: Mutations and Polymorphisms Detected by the xTAG® CYP2D6 Kit v3

Star Genotype (*)	**Mutations and Polymorphisms[+] detected by xTAG® CYP2D6 Kit v3**	
	PCR A	**PCR B**
*1	None	None
*2	**-1584C>G**, 1661G>C	**2850C>T, 4180G>C**
*3		**2549A>del**
*4	100C>T, 1661G>C, **1846G>A**	2850C>T, 4180G>C
*5		**Deletion**
*6	**1707T>del**	4180G>C
*7		**2935A>C**
*8	1661G>C, **1758G>T**	2850C>T, 4180G>C
*9		**2613delAGA**
*10	**100C>T**	1661G>C, 4180G>C
*11	**883G>C**, 1661G>C	2850C>T, 4180G>C
*15	**138insT**	
*17	**1023C>T**, 1661G>C	2850C>T, 4180G>C
*29	**1659G>A**, 1661G>C	2850C>T, **3183G>A**, 4180G>C
*35	-1584C>G, **31G>A**, 1661G>C	2850C>T, 4180G>C
*41	1661G>C	2850C>T, **2988G>A**, 4180G>C
DUP	**Duplication**	

[+]Nucleotide changes that define the star genotype (*) are shown in bold font.

Performance Characteristics (if/when applicable):

Clinical Performance Characteristics:

a) Method Comparison Studies / Accuracy:

Accuracy was assessed through evaluation of samples representing all alleles probed by the assay. The majority of samples consisted of left-over, anonymized, banked extracted DNA from EDTA or citrate anticoagulated whole-blood specimens. For rare alleles, the sample set was supplemented with "blended" samples where linearized plasmid DNA harboring the rare mutations was quantitatively added to genomic DNA to mimic a heterozygous sample when tested. Method comparison studies were performed using bi-directional DNA sequencing as the comparator for the xTAG® CYP2D6 Kit v3. Four hundred and fifty-nine (459) clinical samples were analyzed in the accuracy study. Bidirectional sequence analysis for genotype confirmation was performed for all 459 clinical samples analyzed by the xTAG® CYP2D6 Kit v3.

The overall accuracy of the xTAG® CYP2D6 Kit v3 is 98.47 percent across all mutant and wild-type alleles when compared to bidirectional dideoxy sequencing after the first test. There was one 'No Call' and one incorrect call after the first re-test. For the sample that resulted in the incorrect call, the xTAG® CYP2D6 Kit v3 reported a HET at the 31G>A loci (both a G and A present at the loci), resulting in an overall genotype call of *10/*35 instead of the expected *2/*10 genotype. The overall accuracy was 99.56 percent after the first re-test. The accuracy agreement for genotype detection of the xTAG CYP2D6 Kit v3 was calculated by determining the percentage of tested samples with the correct genotype call, compared to the total number of samples tested of that genotype.

Accuracy for Rare Alleles

Plasmid clones for the *8 allele were created by blending plasmids harboring mutations for rare alleles with genomic DNA. A total of 5 blends of genomic DNA and plasmid were tested. Four replicates of each blend were amplified (total of 20 samples) and assayed by xTAG® CYP2D6 Kit v3. Genotype results using plasmid clone-genomic DNA blends are shown in Table 2. The CYP2D6 genotype call rate was 100 percent for plasmid clone genomic DNA blends.

Table 2 – Accuracy for Rare Alleles

Plasmid Genotype	Genomic DNA Genotype	Genotype of Blended Sample	Blended samples (n)	Number of Correct Calls	Number of Miscalls	Number of No calls	Genotype Call Rate
*8	*1/*1	*1/*8	4	4	0	0	100%
*8	*35/*35	*35/*8	4	4	0	0	100%
*8	*17/*17	*17/*8	4	4	0	0	100%
*8	*2/*2	*2/*8	4	4	0	0	100%
*8	*4/*4	*4/*8	4	4	0	0	100%
TOTAL			**20**	**20**	**0**	**0**	**100%**

b) Reproducibility:

Reproducibility was assessed using a multi-centre, multi-operator, multi-lot, blinded design. A subset of samples were extracted at each site with a different extraction method.

Sample type A: reproducibility of the complete assay, including the extraction step, assessed on 8 clinical (whole blood) samples representing 8 unique genotypes with one wild-type (*1/*1) and one homozygous mutant genotype.

Sample type B: reproducibility of the analytical (post-extraction) steps of the assays, assessed on 5 purified genomic DNAs from cell-lines representing 5 unique mutant genotypes.

The genotypes of the reproducibility sample panel are provided in Table 3.

Table 3 – Reproducibility Sample Panel

CYP2D6 Genotype	Sample Type
*1/*5	extracted DNA
*1/*41	extracted DNA
*4/*35	extracted DNA
*2/*4, DUP	extracted DNA
*35/*41	extracted DNA
*1/*10	whole blood
*10/*17	whole blood
*1/*2	whole blood
*1/*1 (WT)	whole blood
*2/*10	whole blood
*2/*17	whole blood
*1/*5	whole blood
*2/*2 (MUT)	whole blood

DUP, duplication; WT, wild-type; MUT, homozygous mutant.

This study was designed to distinguish between Site-to-Site, Lot-to-Lot and Operator-to-Operator reproducibility in the assay. There were two operators per site, each performing 1 run / day across 3 non-consecutive days (3 runs per operator, or 6 runs per site). Each site used a different extraction method. Each operator across the three independent sites tested identical copies of sample type A and sample type B reproducibility sample sets described above. There were 26 No Calls before allowable re-runs due to operator error. These No Calls were resolved after re-testing. There were no incorrect calls. The overall reproducibility of the xTAG® CYP2D6 Kit v3 was 100 percent after allowable re-runs.

c) Detection Limit and range of assay:

The limit of detection and the input range of the xTAG® CYP2D6 Kit v3 were assessed by analyzing serial dilutions of 4 genomic DNA samples and 1 whole blood sample containing one or more mutations or polymorphisms analyzed by the assay. Serial dilutions of each sample were tested at 9 concentrations (300, 150, 50, 10, 5, 2.5, 1.25, 0.5, and 0.1 ng/µL) in duplicate. Total input DNA was 1800, 900, 300, 60, 30, 15, 7.5, 3 and 0.6 ng. The 4 genomic DNA samples were tested at 8 concentrations (100, 82.5, 50, 10, 5, 2, 1 and 0.1 ng/µL) in duplicate. The whole blood sample was tested at 7 concentrations following extraction (82.5, 50, 10, 5, 2, 1 and 0.1 ng/µL). Forty (40) replicates of each of the 5 samples (4 genomic DNA and 1 whole blood) were run at the above concentrations within the assay range. The range for the xTAG® CYP2D6 Kit v3 was established as 2 ng/µL (Limit of Detection) to 300 ng/µL. The recommended concentration of DNA to be run with the assay is 10 ng/ µL.

d) Analytical Specificity / Interfering Substances:

Elevated levels of hemoglobin, bilirubin, albumin, and tryglycerides in specimens have been shown not to interfere with the performance of the xTAG® CYP2D6 Kit v3.

One of the whole blood samples was wild type for xTAG® CYP2D6 Kit v3 mutations and 5 represented mutations in the xTAG® CYP2D6 Kit v3 panel (*1/*5, *2/*17, *1/*10, *10/*17 heterozygotes and one *2/*2 homozygote). Samples were spiked with the potential inhibitors, then extracted with three commercially available extraction methods and assayed. The levels of interfering substances spiked into the specimens were as follows: Hemoglobin - 500 mg/dL, Albumin – 6000 mg/dL, Bilirubin – 20 mg/dL and Triglycerides – 3000 mg/dL.

Hemoglobin (500 mg/dL), bilirubin (20 mg/dL), albumin (3000 mg/dL) and triglycerides (2000 mg/dL) did not interfere with the performance of the xTAG® CYP2D6 Kit v3.

e) Matrix Comparison (EDTA vs Citrate).

Twenty five (25) independent blood samples from anonymous donors were collected in both citrate and EDTA blood collection tubes. These samples were extracted and then assayed with the xTAG® CYP2D6 Kit v3 in one run.

No differences were observed between the final genotyping calls made for all the samples when the same sample was collected in either EDTA anticoagulant or citrate anticoagulant. There were no reported No Calls.

f) Stability:
The expiration date for xTAG® CYP2D6 Kit v3 will be based on real-time stability testing.

g) Assay Cut-off:
N/A

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food & Drug Administration
10903 New Hampshire Avenue
Building 66
Silver Spring, MD 20993

AUG 26 2010

Luminex Molecular Diagnostics, Inc.
c/o Gloria Lee, Ph.D.
Manager, Regulatory Affairs
439 University Ave.
Toronto, Ontario M5G 1Y8
Canada

Re: k093420
Trade Name: xTAG CYP2D6 Kit v3
Regulation Number: 21 CFR §862.3360
Regulation Name: Drug metabolizing enzyme genotyping system
Regulatory Class: Class II
Product Codes: NTI
Dated: August 20, 2010
Received: August 23, 2010

Dear Dr. Lee:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820).

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801), please contact the Office of *In Vitro* Diagnostic Device Evaluation and Safety at (301) 796-5450. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding postmarket surveillance, please contact CDRH's Office of Surveillance and Biometric's (OSB's) Division of Postmarket Surveillance at (301) 796-5760. For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm for the CDRH's Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 796-5680 or at its Internet address http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm.

Sincerely yours,

Courtney C. Harper, Ph.D.
Director
Division of Chemistry and Toxicology
Office of *In Vitro* Diagnostic Device
Evaluation and Safety
Center for Devices and Radiological Health

Enclosure

K093420

Indication for Use

510(k) Number (if known): k093420

Device Name: xTAG® CYP2D6 Kit v3

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Prescription Use ___X___ And/Or Over the Counter Use ____.
(21 CFR Part 801 Subpart D) (21 CFR Part 801 Subpart C)

(PLEASE DO NOT WRITE BELOW THIS LINE; CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of In Vitro Diagnostic Device Evaluation and Safety (OIVD)

Division Sign-Off
Office of In Vitro Diagnostic Device
Evaluation and Safety

510(k) K093420