INTRODUCTION

Philadelphia (Ph) chromosome is generated through the translocation between chromosomes 9 and 22, and is a hallmark of chronic myeloid leukemia (CML). This translocation results in formation of the BCR-ABL fusion gene. The vast majority of CML patients exhibit breakpoints in two regions; after the 13th exon resulting in the b2a2 (e13a2) fusion or after the 14th exon resulting in the b3a2 (e14a2) fusion. These fusions encode for the P210 onco-protein, which is a constitutively active tyrosine kinase. Molecular detection and quantification of the b2a2/b3a2 fusions are essential for CML diagnosis, monitoring of therapeutic responses and minimal residual disease detection.

KIT CONTENTS

The Clarity™ Major BCR-ABL Mutation Quantification Kit is designed for the detection and quantification of b2a2/b3a2 cDNA on the Clarity™ digital PCR system (Cat. No. 10001). Each kit includes reagents sufficient to perform 96 reactions.

Reagents Supplied *	Volume (µL)
Major BCR-ABL Primer and Probe Mix	66
BCR-ABL dPCR Master Mix (2X)	830
PCR grade Water	440
b2a2 Positive Control^	85
b3a2 Positive Control^	85

^Sufficient for at least 24 reactions
**Reagents for reverse transcription is not included in this kit.*

STORAGE AND STABILITY

The Clarity™ Major BCR-ABL Mutation Quantification Kit should be stored at -20°C upon receipt. Avoid repeated freezing and thawing of kit contents. The kit is stable through the expiry date indicated on the kit label.

EXPERIMENTAL PROCEDURE

1. Thaw reagents at room temperature. When reagents are completely thawed, mix contents by gentle vortexing and centrifuge to collect contents at the bottom of the tubes.

2. Prepare each reaction mix according to the following:

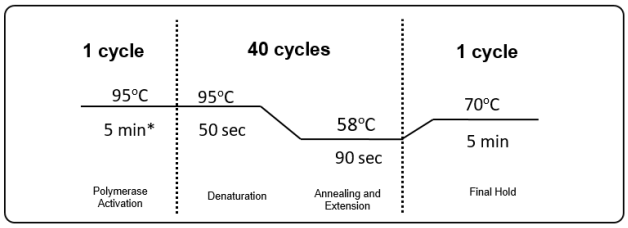
No.	Reagents	Volume (µL)
1	JN Solution (20X)*	0.75
2	Major BCR-ABL Primer and Probe Mix	0.6
3	BCR-ABL dPCR Master Mix (2X)	7.5
4	DNA sample or control	3
5	PCR grade Water	3.15
	Total Vol	15

**Part of Clarity™ 10K consumables package (Cat. No. 10011). Not provided in this kit.*

3. Mix thoroughly by pipetting up and down. Centrifuge to collect contents at the bottom of the tubes.

4. Load sample onto Clarity™ Tube-strips and perform sealing according to instructions provided in the Clarity™ Digital PCR System User Manual.

5. Perform PCR using a deep-well (0.2 ml) thermal cycler using the recommended conditions as shown.



Ramp rate: 1°C/sec

6. Proceed with data acquisition and analysis with default setting for both FAM channel and HEX channel. Refer to the Clarity™ Digital PCR System User Manual for detailed data acquisition and analysis instruction.

For research use only. Not intended for any animal or human therapeutic or diagnostic use.