

Xpert PML-RARA Linearity Panel C221

INTENDED USE:

The Xpert PML-RARA Linearity Panel C221 is intended for use as a reference material to monitor the reportable range of *in vitro* laboratory nucleic acid testing procedures for the quantitative detection of PML-RARA transcript isoform bcr1 and ABL1 endogenous control mRNA transcript when analyzed using the Xpert® PML-RARA assay on Cepheid GeneXpert® Instrument Systems.

The translocation t(15;17) (q24;21) of the promyelocytic leukemia gene (PML) and retinoic acid receptor- α protein (RARA) gene results in PML-RARA, an oncogenic fusion gene. PML-RARA is a driver for acute promyelocytic leukemia (APL), a subtype of acute myeloid leukemia (AML) and is found in over 98% of APL patients². PML-RARA represses the transcription of multiple genes involved in myeloid differentiation and confers a survival and proliferative advantage to leukemic cells, resulting in accumulation of promyelocytes in bone marrow.¹ There are three typical PML-RARA isoforms: bcr1 (L or long), bcr2 (V or variant), and bcr3 (S or short). Bcr1 and bcr3 isoforms are most common and found in 90-95% of APL patients¹. Treatment involving all-trans-retinoic acid (ATRA), arsenic trioxide (ATO), and/or chemotherapy has been largely successful for pediatric and adult patients. Measurable residual disease monitoring (MRD) with qPCR-based detection of PML-RARA transcript is an important tool for use as a prognostic/predictive biomarker to inform treatment decision-making, a monitoring tool to identify impending relapse, and a potential surrogate end point for overall survival in clinical trials to accelerate the development of novel treatment strategies².

PRODUCT SUMMARY and PRINCIPLE:

The Xpert PML-RARA Linearity Panel C221 consists of 5 components. Each component contains an increasing concentration of PML-RARA bcr1 mRNA transcript with fixed concentrations of ABL1 to produce five levels: PML-RARA bcr1 0.1%, PML-RARA bcr1 1%, PML-RARA bcr1 20%, PML-RARA bcr1 100%, and PML-RARA bcr1 450%.

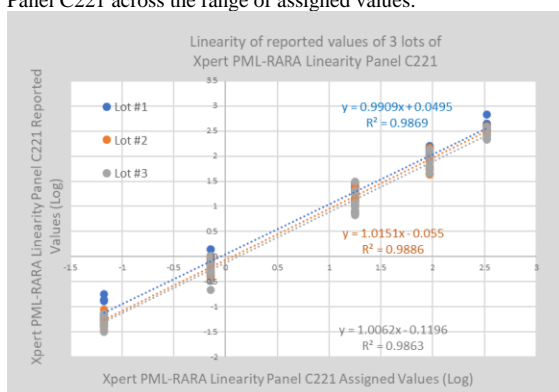
Assessing the linearity of an assay is a crucial part of good laboratory practice, as it confirms that the test system accurately measures patient samples throughout the manufacturer's designated reportable range. Linearity testing can help identify problems related to reagents, sample handling, and instruments, enabling prompt action which may avoid failed assay runs.

Validation and Value Assignment

MMQCI manufactured 3 lots of Xpert PML-RARA Linearity Panel C221 and tested the lots across 3 Xpert PML-RARA assay cartridge lots, incorporating multiple days and operators. Grubb's outlier test was applied to remove statistical outliers and the remaining data was used to assign % values to each level.

Figure 1.

Linearity of the reported values of 3 lots of Xpert PML-RARA Linearity Panel C221 across the range of assigned values.



COMPOSITION:

Xpert PML-RARA Linearity Panel C221 is comprised of 10 single use bottles, 2 bottles of each % PML-RARA level. The C221 bottles contain 4 mL of synthetic RNA transcripts, suspended in a stabilizing matrix with a non-infectious solution of buffers and preservatives. Levels PML-RARA bcr1 0.1%, PML-RARA bcr1 1%, PML-RARA bcr1 20%, PML-RARA bcr1 100% and PML-RARA bcr1 450% contain varying ratios of PML-RARA RNA transcript to ABL1 RNA transcript.

STORAGE and STABILITY:

The Xpert PML-RARA Linearity Panel C221 should be stored at -25°C to -15°C. Unopened material is stable through the expiration date printed on the kit label when consistently stored frozen. Xpert PML-RARA Linearity Panel C221 components are for single use only. Discard after use according to your local and federal regulations.

PRECAUTIONS and WARNINGS:

- Use the Xpert PML-RARA Linearity Panel C221 as provided. Do not dilute or transfer to another storage tube.
- This product is intended for *in vitro* analytical testing and is provided for Research Use Only. It is not for use in diagnostic procedures.
- This product is slightly cloudy in appearance.
- This product does not contain any biological material of human or animal origin. Universal Precautions are NOT required when handling this product.
- Reference materials should be used in accordance with local, state, federal regulations and accreditation requirements.
- Xpert PML-RARA Linearity Panel C221 cannot be cloned, sold, or transferred without the explicit written consent of MMQCI.

INSTRUCTION FOR USE:

1. Allow the Xpert PML-RARA components to be tested to come completely to room temperature (18°C to 25°C) for approximately 30 minutes.
2. Immediately before pipetting, thoroughly mix the control bottle by inverting 8 times followed by 2 pulse vortexes, 2-3 seconds each, at maximum speed.
3. Add 4mL of the control sample to 100 μ L of Proteinase K in a conical tube, as you would a blood specimen.
4. Continue with the assay procedure according to manufacturer's instructions.
5. Discard after use according to local and federal regulations.

EXPECTED VALUES:

Each kit box of Xpert PML-RARA Linearity Panel C221 contains a lot-specific Data Sheet that details the assigned % values for that lot number of C221. Once the laboratory has tested all levels of the C221 panel using the Xpert PML-RARA assay (replicates are recommended for linearity assessment), perform a linear regression analysis to calculate a correlation coefficient (R^2), and graph the data by plotting the reported % values against the assigned values. See Figure 1 of the Data Sheet for an example of a linear regression graph. An R^2 of 0.9 or higher confirms linearity across the reportable range of the assay. ***It is important to note that reported % values may vary among laboratories due to different reagent lots, operators, and test systems. Each laboratory should establish their own % ranges for the C221 panel.***

References

- ¹ Liquori A, Ibañez M, Sargac C, Sanz MÁ, Barragán E, Cervera J. Acute Promyelocytic Leukemia: A Constellation of Molecular Events around a Single PML-RARA Fusion Gene [published correction appears in *Cancers (Basel)*. 2021 Jul 09;13(14):]. *Cancers (Basel)*. 2020;12(3):624.
- ² Heuser M, Freeman SD, Ossenkoppele GJ, et al. 2021 Update on MRD in acute myeloid leukemia: a consensus document from the European LeukemiaNet MRD Working Party. *Blood*. 2021;138(26):2753-2767.

ORDERING INFORMATION:

Xpert PML-RARA Linearity Panel C221

Part Number: C221

Kit contains: 10 bottles x 4mL

2 of each % Level:

Levels: bcr1 0.1%, bcr1 1%, bcr1 20%, bcr1 100% and bcr1 450%