Virongy Biosciences
Accelerate Discovery

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Replication-Competent Lentivirus (RCL) Assay Report

1. Client and Study Information

Client:
Contact:
Study ID:
Report Date:
Sample ID:
Sample Description:

2. Assay Summary

The purpose of this assay was to determine whether replication-competent lentivirus (RCL) is present in the submitted lentiviral vector preparation. The assay utilizes an amplification culture system using the C8166 T-cell line, which is permissive for replication-competent lentivirus. Test samples were inoculated onto C8166 cells and maintained in culture for a period of 14 days to allow amplification of any replication-competent lentivirus that may be present. Cell cultures were monitored throughout the incubation period and maintained under standard growth conditions with periodic media replacement and cell passaging as required. A replication-competent lentivirus derived from the R8.71 packaging construct was used as the positive control to confirm assay sensitivity and the ability of the culture system to support viral replication. Negative control cultures were maintained in parallel without viral inoculation. At the conclusion of the two-week amplification period, culture supernatants were collected and analyzed for evidence of replication-competent lentivirus. The performance of the positive and negative controls was evaluated to verify that the assay met the required acceptance criteria.

3. Sample Information

Sample ID	Lot Number	Matrix	Volume Received	Storage
LV-001	Lot-23045	Viral Supernatant	2 mL	-80°C

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4. Controls

Control	Expected Result	Observed Result
Positive Control R8.71	Detected	Detected
Negative Control LentiGFP(VSVG)	Not Detected	Not Detected
No Template Control	No Amplification	No Amplification

5. Amplification Culture Timeline

Passage	Day	Observation
P0	Day 0	Cells inoculated with test sample
P1	Day 7	Cells passaged
P2	Day 14	Supernatant collected for qPCR

6. qPCR Detection Results

Sample	Target	Ct Value	Result
Negative	gag	ND	Negative
Positive Control	gag	23.4	Positive

7. Limit of Detection

The validated limit of detection for this assay is approximately 10 infectious replication-competent lentivirus particles per sample following amplification.

8. Conclusion

No replication-competent lentivirus (RCL) was detected in the tested sample under the conditions of this assay. The sample meets the acceptance criteria for RCL testing.

9. Signatures

Role	Name	Date
Technician		
Reviewed By		