

Resource Summary Report

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ddPCR Titration of Lentivirus Vectors

DOI:10.17504/protocols.io.be7ijhke

Type: Protocol

Proper Citation

Addgene The Nonprofit Plasmid Repository 2020. ddPCR Titration of Lentivirus Vectors. protocols.io dx.doi.org/10.17504/protocols.io.be7ijhke

Protocol Information

URL: <https://dx.doi.org/10.17504/protocols.io.be7ijhke>

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Summary: This protocol describes ddPCR titration of Lentivirus vectors. To see the full abstract and additional resources, visit <https://www.addgene.org/protocols/lentivirus-ddpcr-titration/>. This protocol was modified from the publication Wang Y, Bergelson S, Feschenko M, 2018. Sample Data: When analyzing data there should be a clear distinction between negative droplets (black) and positive droplets (blue/green). The concentration of RRE positive droplets in the untransduced control should be close to zero (A01). In this protocol, the lentiviral particles are serially diluted and used to transduce HEK293T cells. Genomic DNA is extracted from the target cells and assayed for integrated copies of RRE. Since the samples that are assayed are diluted 2-fold serially, the concentration of RRE positive droplets should decrease by a factor of 2 across the dilutions. RPP30 copies should be relatively constant across samples. In the RRE example below, 2-fold serial dilutions of a sample were loaded in wells B01-H01. As shown in the image and table below, the concentration of RRE positive droplets increases by a factor of ~2 as you progress from the higher dilutions to the lower dilutions (blue). The concentration of RPP30 positive droplets stays relatively even across samples (green). To increase the accuracy of the titer, calculate and average of several dilutions.

Affiliations: Addgene

External URL: <https://www.addgene.org/protocols/lentivirus-ddpcr-titration/>

Version: 1

Publication Date: 2020

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Record Creation Time: 20210329T220522+0000

Record Last Update: 20210329T220856+0000

Data and Source Information

Source: