# Certificate of Analysis



# Guide-it™ Genome-Wide sgRNA Library Transfection Mix

Catalog No. Amount Lot Number

632650 (Not sold separately) Sold as a part of 632646 2 x 5 Vials Specified on product label

# **Description**

The Guide-it Genome-Wide sgRNA Library Transfection Mix is sold as part of our Guide-it CRISPR Genome-Wide sgRNA Library System for performing phenotypic knockout screens in human cells. There is no need to amplify the library, as each vial contains all the necessary plasmids and transfection reagents to generate self-inactivating lentiviral particles containing a complete sgRNA library targeting the entire human genome (19,000 genes). Simply resuspend the transfection mix in water and add the mixture to Lenti-X<sup>TM</sup> 293T cells to produce lentiviral particles. The sgRNA library is expressed from the U6 promoter and contains four highly active guides per gene, with guide design based on the Brunello library (Doench et al. 2016). The library backbone is the pLVXS-sgRNA-mCherry-hyg Vector, which is also included in our library system. In addition to expressing sgRNA, the sgRNA lentiviral particles also code for mCherry fluorescent protein to enable simple functional titer assays, as well as hygromycin resistance to enable selection for stably transduced cells. To improve the power of your screens, representation of the guide RNAs has been verified in both the plasmid library as well as in transduced target cell populations to be >90% within a 10-fold range.

# **Package Contents**

• 2 x 5 Vials Guide-it Lentiviral sgRNA Library Transfection Mix (Genome-Wide)

#### Storage Conditions

- Store all components at -20°C upon receipt.
- Reconstitute each vial in tissue-culture-grade water immediately before use.

#### **Shelf Life**

• 1 year from date of receipt under proper storage conditions.

# **Shipping Conditions**

• Dry ice  $(-70^{\circ}\text{C})$ 

## **Product Documents**

Documents for our products are available for download at <u>takarabio.com/manuals</u> The following documents apply to this product:

• Guide-it CRISPR Genome-Wide sgRNA Library System User Manual

#### References

Doench, J et al., Optimized sgRNA design to maximize activity and minimize off-target effects of CRISPR-Cas9. *Nat Biotechnol.* **34**, 184–191 (2016).

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# **Quality Control Data**

## **Transfection Test**

Lenti-X 293T cells were transfected with plasmid DNA encoding *AcGFP1* as described in the Lenti-X Packaging Single Shots (VSV-G) Protocol-at-a-Glance. Forty-eight hours post-transfection, AcGFP1 expression was assessed by flow cytometry, and the transfection efficiency (as determined by the percentage of 293T cells expressing AcGFP1) was demonstrated to be >75%. The presence of virus particles in the supernatant of transfected cells was confirmed using Lenti-X GoStix<sup>TM</sup> Plus.

## **Guide RNA Representation**

For each lot, guide RNA representation for all stated sgRNAs within the Guide-it sgRNA Genome-Wide Library was confirmed by NGS to be >90% within a 10-fold distribution range. A complete list of all sgRNAs in our library and their representation data can be found on our website at <u>takarabio.com/genomewidelibrary</u>.

## **Sterility Test**

There was no evidence of bacterial or fungal growth after inoculation on thioglycollate medium and tryptic soy broth.

It is certified that this product meets the above specifications, as reviewed and approved by the Quality Department.

(112718) Page 2 of 2



# Guide-it™ Genome-Wide sgRNA Library Transfection Mix

## CATALOG NO.

632650

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11/18/2020

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