bio-techne® | R&D SYSTEMS

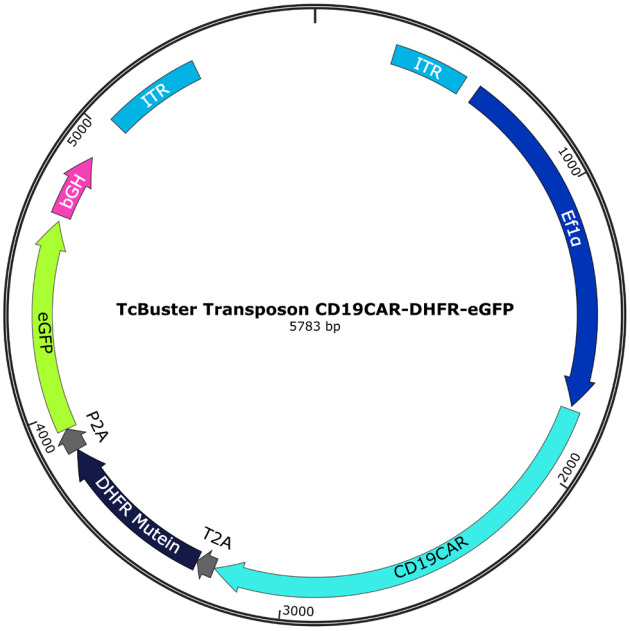
Catalog Number TCBP001-100

Mass 100 µg

TcBuster™ Transposon CD19CAR-DHFR-eGFP

Product Description

Gene	CD19CAR-DHFR-eGFP
Promoter	Ef1α
Transposon Insert Size	5100 bp
Plasmid size	5783 bp
Backbone	Nanoplasmid™
Concentration	100 µg at 2 mg/mL



Preparation and Storage

Formulation	Nuclease free water
Shipping	Ships at ≤ -20 °C.
Storage	Store at ≤ -20 °C. Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

Limitations

- For laboratory research use only. Not for use in diagnostic procedures.
- For certain applications, results may vary due to differences in donor derived cell populations.

Required Reagents and Equipment

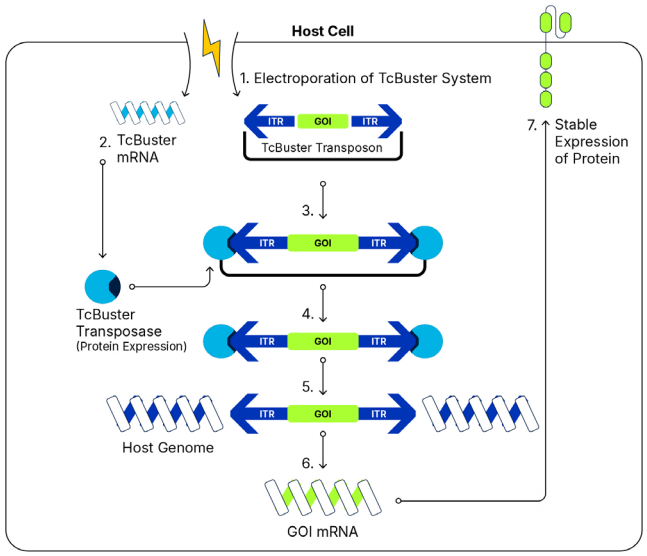
- Electroporation platform of user's choice
- TcBuster-M™ Transposase mRNA, Catalog # TCB-001.1-100 or TCB-001.1-500

Intended Use

TcBuster Transposon CD19CAR-DHFR-eGFP is one part of the TcBuster system and is compatible with TcBuster transposases. Ensure that you have TcBuster-M mRNA (Catalog #: TCB-001.1-100, TCB-001.1-500) prior to using this product. It is highly recommended to deliver TcBuster system reagents (transposase mRNA and compatible DNA transposon) with an electroporation platform. The TcBuster system is a versatile genome editing tool and has applications in a wide variety of cell types. Examples include, but are not limited to, the transposition of immune cells (T, NK), induced pluripotent stem cells (iPSCs), and bioproduction cell lines (CHO, HEK, etc.). Broad application will require independent experimental optimization and/or process development. If you have any technical questions about the TcBuster non-viral gene delivery system or would like to inquire about custom transposons, contact our TcBuster scientific support team at techsupport@bio-techne.com for assistance.

FIGURE // 01

The TcBuster™ System Mechanistic Diagram



1. TcBuster system reagents are introduced into the cells via electroporation.

2. TcBuster-M mRNA is translated into TcBuster-M transposase enzyme.

3. TcBuster-M transposase binds to inverted terminal repeats (ITRs) on the TcBuster DNA transposon.

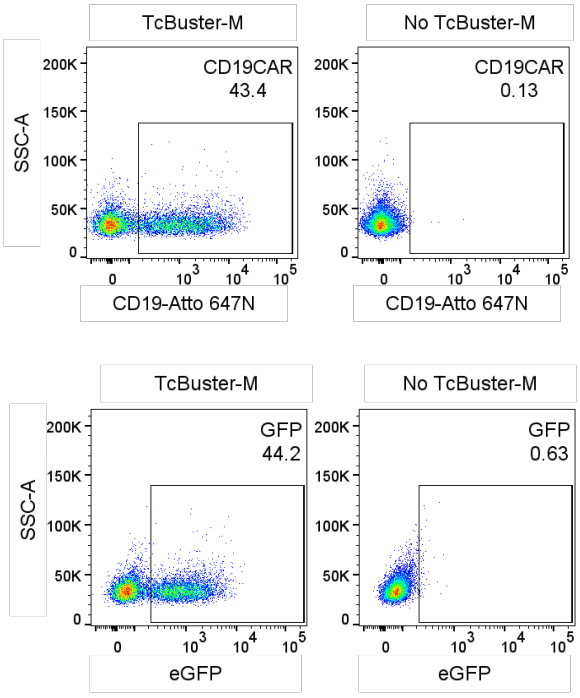
4. TcBuster-M transposase cuts gene(s) of interest (GOI) from transposon.

5. TcBuster-M inserts GOI into the host genome.

6. Inserted GOI is transcribed into mRNA.

7. Translated GOI protein is stably expressed in cells.

Data Images



Primary human T cells edited with the TcBuster system express CD19CAR and eGFP. Primary human T cells were gene edited with TcBuster-M (Catalog # TCB-001.1) transposase and CD19CAR-DHFR-eGFP (Catalog # TCBP001-100) transposon. On day 7 after gene editing, T cells were stained with Recombinant Human CD19 Protein, Atto 647N Conjugate (Catalog # ATM9269) and analyzed by flow cytometry for CD19CAR and eGFP expression. Representative flow cytometry plots of primary T cells edited with or without TcBuster-M transposase.

Background

The TcBuster system is a non-viral gene delivery system that enables stable gene transfer in most cell types. The TcBuster system belongs to the hAT-family of DNA transposons and is derived from the red flour beetle *Tribolium castaneum* (1). The system consists of the TcBuster-M transposase mRNA, which encodes for a hyperactive version of TcBuster transposase, and DNA transposon encoding multicistronic cargos for gene insertion (2). Figure 1 shows a Mechanistic Diagram of how the TcBuster system introduces genetic material into cells. The Ef1α promoter sequence is commonly used to drive gene expression in mammalian cells. This promoter is known for its strong and constitutive activity and is often chosen for use in genetic engineering applications (3). CD19 CAR is a chimeric antigen receptor that is designed to target CD19, a protein that is expressed on the surface of B cells. This receptor has been used in genetically engineered T cell immunotherapies to treat various types of cancer, such as B cell leukemias and lymphomas (4). DHFR, dihydrofolate reductase, is an enzyme that is involved in the synthesis of nucleotides. Muteins of this enzyme has been used as a selectable marker in genetic engineering, allowing for the selection of genetically modified cells using methotrexate (5). eGFP is a variant of green fluorescent protein that is commonly used as a reporter gene in genetic engineering. This protein emits a bright green fluorescence when exposed to blue light, making it a useful tool for visualizing gene expression after genetic modification (6). bGH, or bovine growth hormone polyadenylation signal, is a common terminator in mammalian expression vectors. This sequence mediates efficient and accurate termination of the transcript (7). This product is built with a Nanoplasmid backbone. This technology requires the use of a proprietary strain of bacteria for growth.

Product Specific Notes

Transfer of TcBuster Transposon CD19CAR-DHFR-eGFP (“Product”) is strictly prohibited. The Product is to be used for research use only, and not for clinical or commercial purposes. Full details of R&D Systems' Terms and Conditions of sale can be found online at: https://www.bio-techne.com/terms-and-conditions.

REFERENCES

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