2020 Rare Disease Challenge

Treatment of Spinal Muscular Atrophy Rare Disease With CRISPR Technology

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ABSTRACT

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The biggest development in the biological sciences is undoubtedly the discovery of the DNA double helix. This discovery in 1953 laid the groundwork for the study of structural changes in DNA, and with the development of technology, perhaps this structure will be a beacon of hope for most cancer diseases and rare diseases for humanity to quickly unravel in periods of progress..Polymerases, ligases, restriction enzymes, helicases and polymerase chain reaction (PCR) with the discovery of structures such as the isolation of genes and gene fragments, cell culture, model organisms and in vitro studies of gene mutations and gene maps are of great importance. With the advent of genomic sequencing, genome maps of humans and other organisms began to form. The Clustered Regularly Interspaced Palindromic repeats (CRISPR)-Cas system, a genome sequencing tool based on the principles of the Watson - Crick DNA model, has been of considerable importance to biological science. The CRISPR-Cas system has become a beacon of hope for humanity, especially for the

treatment of rare diseases. In our study, it was aimed to repair the tissue associated with cutting the faulty area with the enzyme Nhae1 restriction over exon 7 using the CRISPR-Cas9 system for Spinal muscular atrophy

disease and adding a properly prepared primer to the cut area. This mechanism is intended to minimize the toxicological effect of herpes viruses and proteins in the preparation and transfer into the cell.

Keywords: Genome, Gene fragments, Palindromic repeats,Spinal Muscular Atrofi

RESULTS

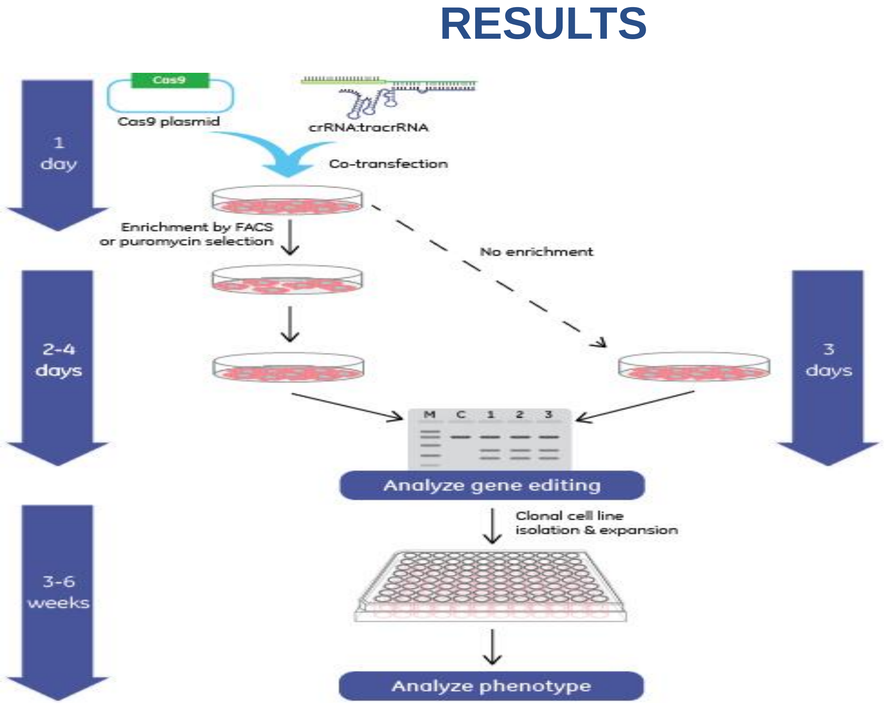

Figure:Gene knockout workflow using Cas9 expression plasmid and synthetic crRNA: tracrRNA

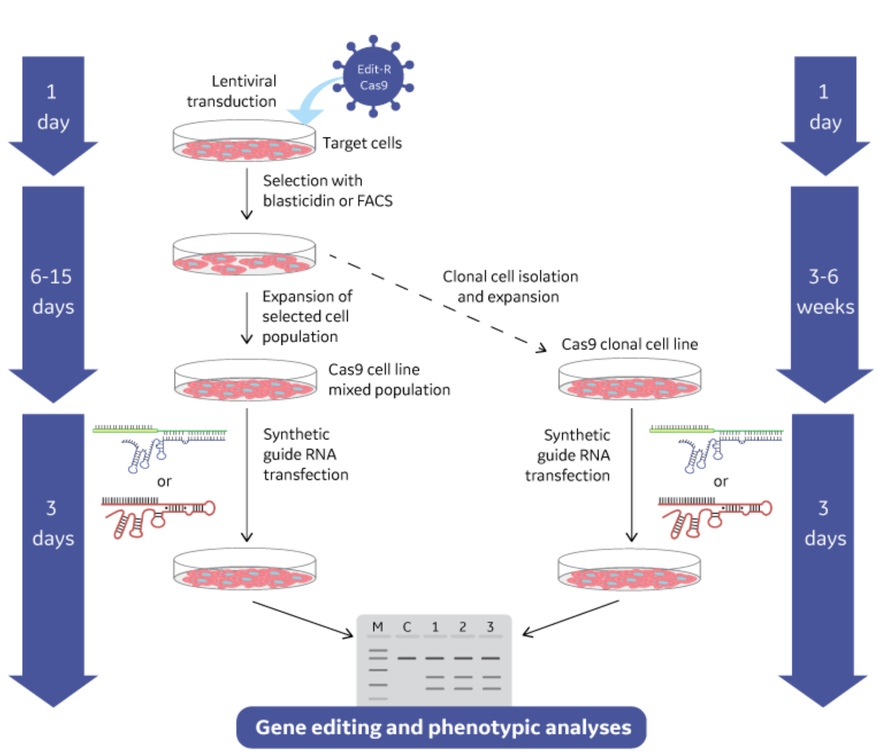

Figure: The structure should be provided as a gene knockout workflow using lentiviral Cas9 expression particles and synthetic crRNA: tracrRNA

CONCLUSION

Stabilizing modifications on synthetic guide RNAs (crRNA:tracrRNA or sgRNA) to resist nuclease degradation are required for co-electroporation with Cas9 mRNA.

Modifications can improve gene editing when

electroporated as RNPs, but this may be targetsequence specific and not a universal benefit.

Modified and unmodified synthetic guide RNAs show no difference in gene activity when delivered by lipid transfection reagents into a Cas9 stable cell line.

Some modification patterns cause increased cellular toxicity

Modification of the single-stranded regions of the guide RNAs modestly increases gene editing in lipid co-transfection with Cas9 mRNA or Cas9

protein while also stabilizing the RNAs for coelectroporation with Cas9 mRNA.

REFERENCES

- Dayangaç D, Erdem Yurter H. RNA “splicing” hataları sonucu ortaya çıkan kalıtsal hastalıklar. Hacettepe Tıp Dergisi 2005; 36:9-12.
- John A. Schiel, Eldon T. Chou, Maren Mayer, Emily M. Anderson , and Anja van Brabant Smith Dharmacon, Homology-directed repair with Dharmacon Edit-R CRISPR-Cas9 and single-stranded DNA oligos. A Horizon Discovery Group Company, Lafayette, CO, USA
- Kaizhang HE. Eldon Chou, Armanda Haas, Zakilna Strezoska, Mellisa L., Kelley and Anja van Braband Smith. CRISPR/Cas9 genome editing utilizing chemically synthetsized RNA. GE Healthcare 2650.
- Tiana Stastny, Megan Basila, Hidevaldo B. Machado, Emily Anderson, Eldon T. Chou, Melissa L. Kelley, Anja van Brabant Smith Dharmacon, 2’-O-methyl phosphorothioate linkagemodified synthetic guide RNAs for efficient CRISPR-Cas9 genome editing and reduced cellular toxicity A Horizon Discovery Group Company, Lafayette, CO, USA

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