RaDiChal'21
FINAL

2021 Rare Disease Challenge

PRIME EDITING BASED GENE THERAPY FOR FAMILIAL MEDITERRANEAN FEVER VIA EX VIVO LENTIVIRAL DELIVERY TO HEMATOPOIETIC STEM CELLS

TEAM GENIUS

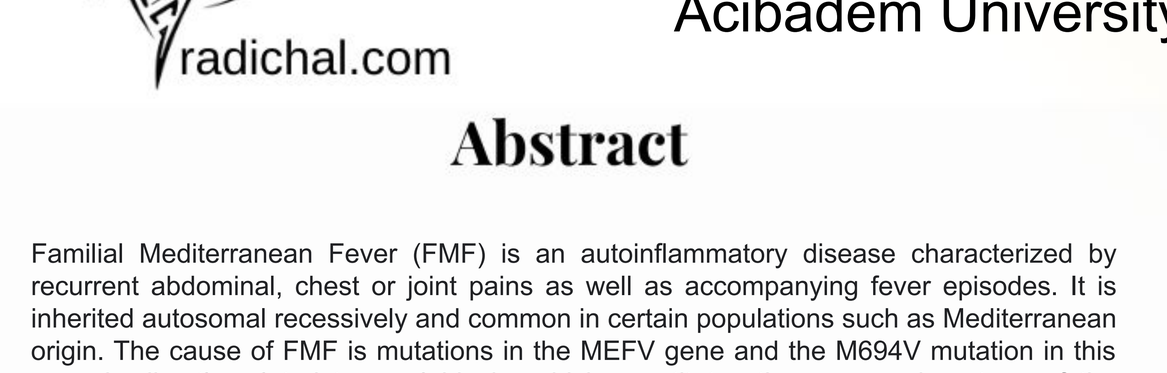
Beril Ay, Efe Sarı, Seray Başak Bozkurt, Yaren Nur Demir,
Acibadem University School of Medicine

Abstract

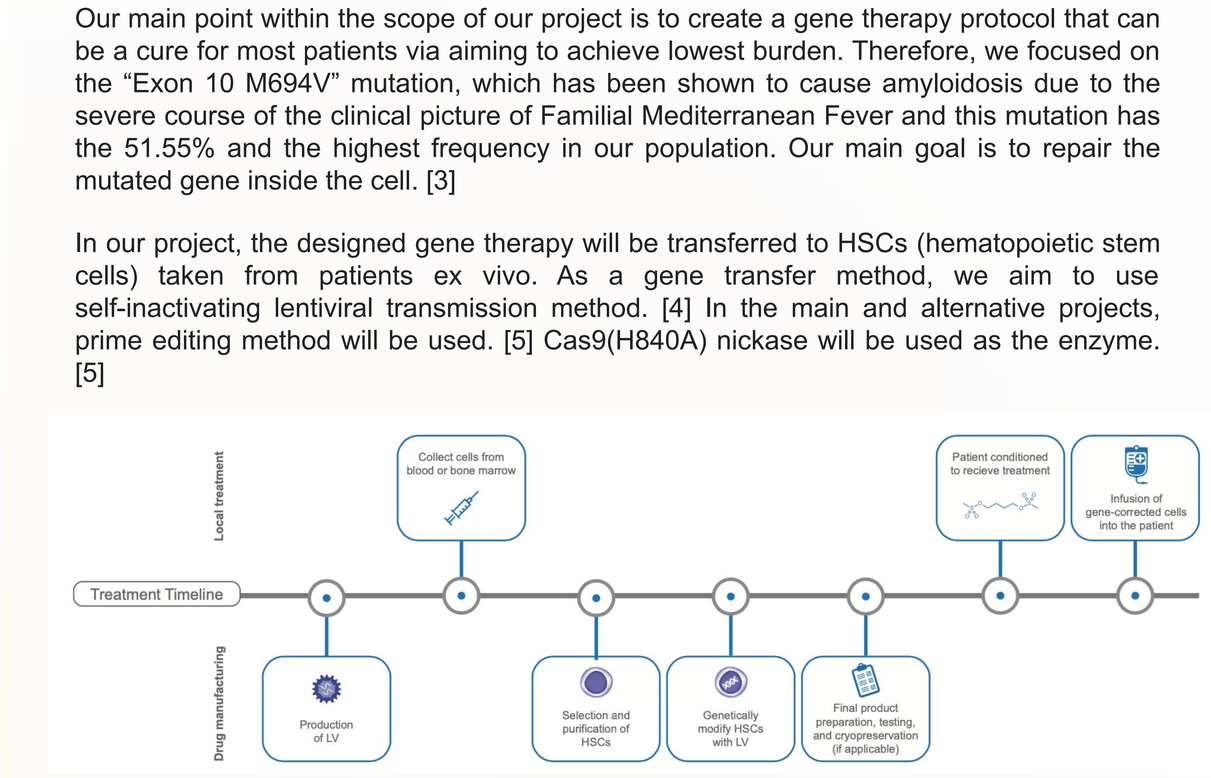
Familial Mediterranean Fever (FMF) is an autoinflammatory disease characterized by recurrent abdominal, chest or joint pains as well as accompanying fever episodes. It is inherited autosomal recessively and common in certain populations such as Mediterranean origin. The cause of FMF is mutations in the MEFV gene and the M694V mutation in this gene is directly related to amyloidosis, which constitutes the severe phenotype of the disease. [1, 2]

Our main point within the scope of our project is to create a gene therapy protocol that can be a cure for most patients via aiming to achieve lowest burden. Therefore, we focused on the “Exon 10 M694V” mutation, which has been shown to cause amyloidosis due to the severe course of the clinical picture of Familial Mediterranean Fever and this mutation has the 51.55% and the highest frequency in our population. Our main goal is to repair the mutated gene inside the cell. [3]

In our project, the designed gene therapy will be transferred to HSCs (hematopoietic stem cells) taken from patients ex vivo. As a gene transfer method, we aim to use self-inactivating lentiviral transmission method. [4] In the main and alternative projects, prime editing method will be used. [5] Cas9(H840A) nickase will be used as the enzyme. [5]



Projects



In our main project, we planned to edit the M694V mutation, which is the most common (51.55%) and associated with the most severe type in patients, with the CRISPR Prime Editing method.

The enzyme planned to be used for this editing process was determined as Cas9 (H840A) Nickase, and the Prime Editing Guide RNA (pegRNA) is designed with a low off-target and high on-target score via the pegFinder program. [6]

The created Prime Editing complex will be made by means of self-inactivated lentiviruses; which can integrate into the genome of the target cell and have an advantageous use to maintain a long and desired level of gene expression in rapidly dividing cells such as hematopoietic stem cells by deleting 400 nucleotides from the LTR region. [7]

According to our gene therapy protocol, firstly, hematopoietic stem cells with increased mobility in the blood will be collected from patients by apheresis, then, after gene editing as mentioned above, they will be transferred back to the patient with myeloablative conditioning via intravenous route and will be expected to settle in the bone marrow and eventually, this will be a solution to our patient's clinical outcomes.

Social Impact

@geniusteam21 Followers: 201 Likes: 672
Sharings: 24 Watchs: 165

Informative posts and stories

An interview with Prof.Dr. Özgür Kasapçopur:

- The interview was sent to UluBAT (National Scientific Research Society), which aims to provide scientific research infrastructure for students which reaches around 30,000-40,000 medical students.

The article “Chaos to Diagnosis: FMF”:

- published in Euromeds Blog, the official publication of EMSA (European Medical Students' Association) Europe, which brings together over 50,000 medical school students in more than 30 countries.

Flyer distribution and street interview in Kadikoy

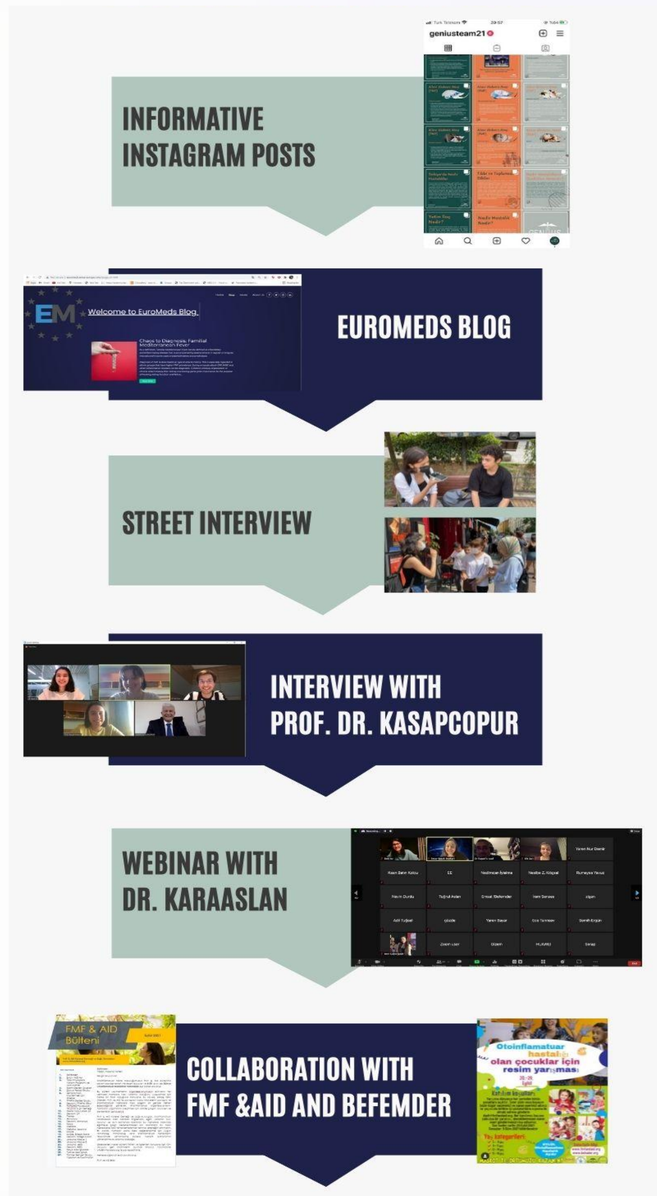
A webinar on FMF and rheumatological diseases with Dr. Mehmet Karaaslan, Acibadem Altunizade Hospital Rheumatology Department physician

Collaboration with FMF & AID Global Association for FMF & AID Newsletter:

- We translated the newsletter published by the FMF&AID Association on the 17 September FMF Day into Turkish.

Painting Contest for Children with Autoinflammatory Disease:

- We organized a painting competition with international cooperation.



Conclusion

All in all, as Genius Team, with the gene therapy protocol we designed, we aim to find a cure for FMF patients experiencing clinical symptoms, especially the most severe one, amyloidosis. In the main and alternative projects, we planned an ex vivo therapy protocol, in which autologous hematopoietic stem cell transplantation will be conducted. In these projects, we opt to design and correct the missense mutation (M694V) located on the exon 10 of the MEFV gene present in chromosome 16. To achieve this, we plan to use CRISPR Prime Editing Technology and transfer our editing complex to hematopoietic stem cells of FMF patients via lentiviral transfer vectors ex vivo.

References