Bachelor Thesis
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Novel gene therapy approach combined with anti-TLR4 monoclonal antibodies in MPS VII.

Universitat Autònoma de Barcelona

Degree in Biochemestry - Universidad Autónoma de Barcelona
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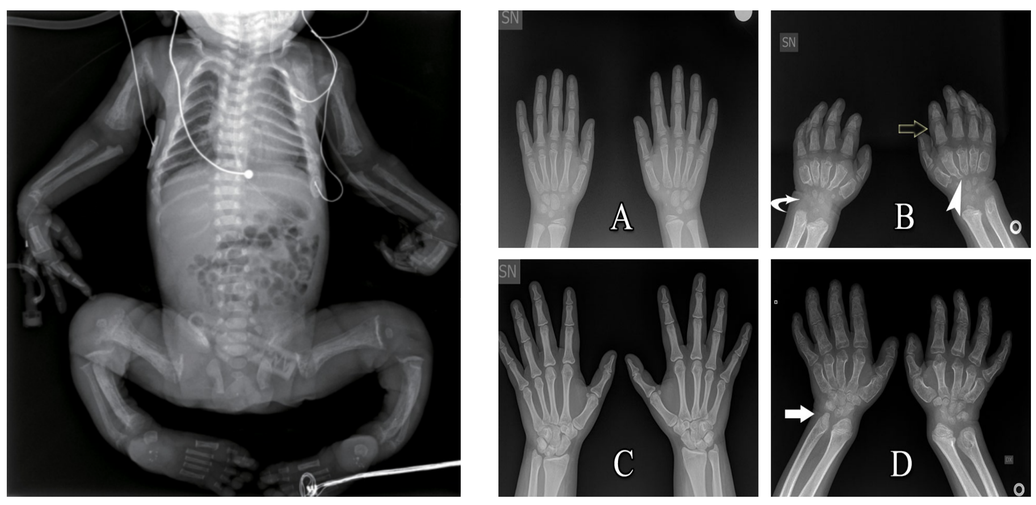
Introduction

The mucopolysaccharidosis (MPS) are a subset of lysosomal storage disorders characterized by deficiencies in enzymes that contribute to degradation of glycosaminoglycans (GAGs). GAGs accumulate in the lysosome, leading to dysfunction in a variety of cell types and organs through mechanisms that generally remain unclear. MPS are considered rare diseases due to its low incidence, which is estimated in 1 of 25.000 live births. Concretely, mucopolysaccharidosis type VII (MPS VII) is an autosomal recessive disorder caused by a severe deficiency in the activity of the lysosomal enzyme β-glucuronidase, an enzyme involved in the stepwise degradation of GAGs.

Symptoms

MPS patients exhibit multi-systemic disease symptoms like hepatosplenomegaly, corneal clouding, growth and mental retardation, cardiac defects, and bone malformations collectively known as dysostosis multiplex.



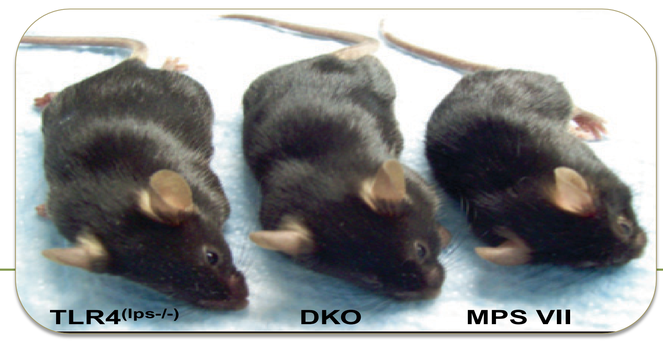
Skeletal pathology becomes progressively worse with age, bones develop poorly, are thick, short and angulated, leading to disproportional short stature.

Objective

The main objective is to propose a PhD thesis that consists in a gene therapy approach, which tries to revert mucopolysaccharidosis type VII clinical features, concretely bone and joint complications.

Methodology

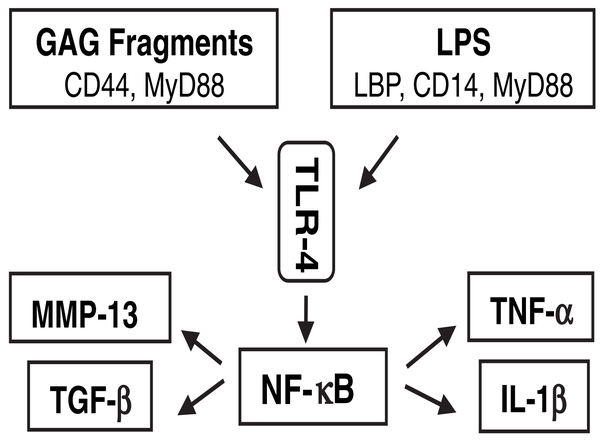
- Preliminary information of MPS VII pathogenesis and complications was collected from articles and web sites (including MPS Spain Association).
- In order to not overlap previous knowledge and trying to propose a novel approach, different projects were consulted. Besides, a virtual interview with the neuropediatrician Dr. Mercedes Pineda Marfa from Sant Joan de Déu Hospital (Barcelona) was done.

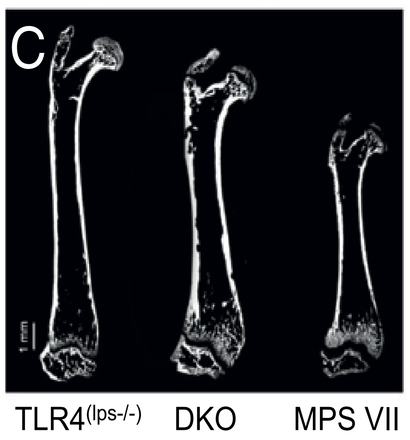


TLR4 pathway

Up regulation of destructive enzymes in MPS could induce inflammatory pathways and altered growth of connective tissue and other cells through activation of the Toll-like receptor 4 (TLR4) signalling pathway. Liposaccharide (LPS) is the classical ligand of TLR4 but heparan sulfate, one of the GAGs that accumulates in MPS VII, acts as an endogenous ligand of TLR4 and activates an inflammatory response via the NFκB signalling pathway.

Simonaro et al., with the aim to examine the pathophysiological significance of TLR4 activation in the MPS disorders, generated double-KO (DKO) mice as a result of crossing TLR4 (LPS−/−) mice with MPS VII mice.





Hypothesis

The main problem is the delivery of the vector into bone and joints due to they are compact and poorly irrigated tissues. Besides, it is supposed to be an activity increment in the TLR4 pathway of MPS VII patients that causes an enhanced expression of NF-kB and a set of pro-inflammatory cytokines. This local increment of pro-inflammatory implies a greater presence of antibodies and blocking agents that avoid, even more, the vector arrival to these cell types and also induces apoptosis to chondrocytes of the growth plate. **Therefore, my proposal is a gene therapy approach in which, a lentiviral virus that carries a ubiquous and strong promoter (EF1α) upstream of cDNA of GUSB combined with the injection of anti-TLR4 monoclonal antibodies in order to facilitate vector infection in bone and joints**. In this PhD research proposal I propose two main trials:

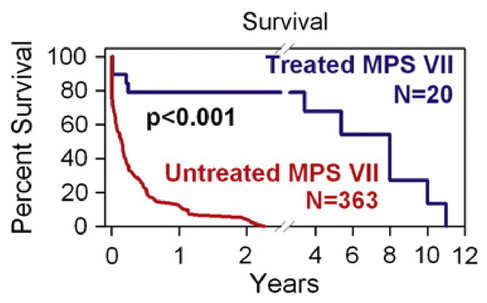
1) A gene therapy approach consisting of a single intravenous systemic injection of lentiviral-vector in MPS VII dogs at birth, combined with intravenous injection* of anti-TLR4 monoclonal antibodies.

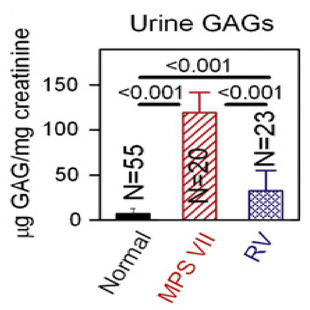
2) A gene therapy approach consisting of single intravenous injection of lentiviral-vector in MPS VII 3-weeks-old dogs after several and continuous join-injections* in synovial fluid of anti-TLR4 monoclonal antibodies during development and mature adults dogs.

*In both trials, anti-TLR4 monoclonal antibodies injections will be done in front and rear right legs and front and rear left legs will be use as a control.

Expected Results

- Anti-TLR4 monoclonal antibodies administration will improve the vector tropism in bones and joints.
- β-glucuronidase activity and GAGs accumulation in the main tissues will be restored. Also in chondrocytes and osteoblasts that will mean longer and shapely bones
- MPS VII treated dogs will have longer survival expectancy than untreated MPS VII dogs.





References

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