The HLA-transgenic mouse as a model system for celiac disease therapy development

Master's Thesis - University of Turku, MSc Degree Programme in Drug Discovery and Development

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Abstract

Background: Celiac disease (CD) is a T helper 1 (Th1)-driven autoimmune condition affecting around 1% of the population worldwide. The only available treatment is a lifelong adherence to a strict gluten-free diet. A significant obstacle to therapy development is the lack of an animal model that fully represents celiac disease pathology. One approach is the usage of mice carrying CD-associated HLA class II transgenes. In this study, the HLA-DQ8, huCD4 transgenic Ab0 (mouse MHC II deficient) NOD mouse is used as a model system for efficacy studies of an immunomodulatory nanoparticle-based treatment for CD. In addition, 2 HLA-DQ2 transgenic mouse lines are being established, to provide cell donor/recipient strains for a humanized gliadin memory T cell adoptive transfer model. Finally, a gluten-containing growth medium is developed, to enable studies of bacterial gluten metabolism.

Methods: 1) 49 mice were divided into three groups. The main group was treated with TIMP-GLIA, a nanoparticle containing the CD antigen gliadin encapsulated in a biodegradable polymer. Treatment was compared to TIMP-OVA, a control nanoparticle loaded with mock antigen. Two groups of mice were pretreated at day -11 and -3, and were then immunized against gliadin on day 1 and day 14. The third group remained naïve to gliadin (negative control). All mice were sacrificed on day 28, and spleens and sera were collected for analysis. Group differences for serum titers of anti-gliadin IgG1 and IgG2c, and proliferation and cytokine secretion of re-stimulated spleen cells were analyzed. 2) For the establishment of 2 HLA-DQ2 transgenic mouse lines, breeding pairs were selected based on FACS phenotyping of immunological markers and crossbred to Rag1-/- mice. 3) Gluten-containing solid and liquid media were developed, the latter using pH-adjusted high-gluten wheat beer as starting material.

Results: 1) Treatment with TIMP-GLIA induced a significant reduction of proliferation and inflammatory cytokine IL-17, IFNγ, and IL-2 secretion by gliadin re-stimulated spleen cells. Additionally, TIMP-GLIA decreased the serum titers of Th1-associated/complement-activating anti-gliadin IgG2c. 2) Mice homozygous for HLA-DQ2 and carrying either a huCD4 transgene or the Rag1 knockout mutation were obtained by crossbreeding. 3) Gluten-containing media were used to test growth of bacterial test species.

Conclusion: 1) Reductions in proliferation, proinflammatory cytokines secretion and anti-gliadin IgG2c suggest that TIMP-GLIA prevents Th1 polarization in gliadin-sensitized HLA-DQ8 mice. 2) Immunocompetent and lymphopenic HLA-DQ2 transgenic mice may be used to develop a humanized gliadin memory T cell adoptive transfer model of CD. 3) Growth of bacterial test species in gluten-containing media proved successful