

# Myofibroblasts and polyploid cells in the conjunctival surface after allogeneic hematopoietic stem cell transplantation

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Av

David Hallberg

Fakultetsopponent: Professor Alexandros Spyridonidis,  
University Hospital of Patras, Patras, Grekland

The thesis is based on the following papers:

- I. **Donor-derived myofibroblasts in the ocular surface after allogeneic haematopoietic stem cell transplantation.**  
Hallberg D, Wernstedt P, Hanson C, Wettergren Y, Stenberg K, Brune M, Stenevi U.  
*Acta Ophthalmol. Scand.* 2006; **84**: 774-780.
- II. **Myofibroblasts in the normal conjunctival surface.**  
Aguilar X, Hallberg D, Sundelin K, Hanson C, Stenberg K, Brune M, Stenevi U.  
*Acta Ophthalmol.* 2010; 88: 407-412.
- III. **Conjunctival polyploid cells and donor-derived myofibroblasts in ocular Graft-versus-Host Disease.**  
Hallberg D, Stenberg K, Hanson C, Stenevi U, Brune M.  
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# Myofibroblasts and polyploid cells in the conjunctival surface after allogeneic hematopoietic stem cell transplantation

David Hallberg

Dept. of Hematology and Coagulation, Inst. of Internal Medicine, University of Gothenburg, Gothenburg, Sweden, 2011.

## ABSTRACT

Allogeneic hematopoietic stem cell transplantation (allo-HSCT) is a potentially curative treatment modality for hematological malignancies, such as leukemia and lymphoma. However, a major complication of the procedure is that the immune effector cells in the graft may become activated towards the host's healthy tissues, resulting in a condition called Graft-versus-Host Disease (GvHD).

Ocular symptoms are among the most common manifestations of GvHD, afflicting at least 50% of all allo-HSCT recipients, typically through dry eye syndrome (keratoconjunctivitis sicca) with features of fibrosis.

This thesis describes a series of studies in which impression cytology was used to sample the ocular surface, of allo-HSCT recipients and healthy individuals, in order to identify and quantify two cell types – myofibroblasts (MFB) and polyploid cells (PP), both of which may play a role in the pathogenesis of ocular GvHD.

Myofibroblasts were identified through immunofluorescence staining for alpha-smooth muscle actin ( $\alpha$ SMA). Moreover, in female allo-HSCT recipients with a male donor, cells of donor origin could be detected through sex chromosome-specific fluorescence-in situ-hybridization (FISH). FISH was also used to identify polyploid cells through their abnormally high chromosome content.

Our results indicate that myofibroblasts of donor origin are present, and increased with time, in the conjunctival surface after allo-HSCT. However, also recipient-derived myofibroblasts was a consistent finding, and were detectable many years after transplantation. These data indicate an ongoing concurrent myofibroblast differentiation from donor and recipient progenitors after allo-HSCT. The donor-derived *versus* total MFB ratio correlated significantly with ocular GvHD. Polyploid cell density increased soon after transplantation, peaked in the 3-12 month interval, and then disappeared.

Compared to post-transplant findings, normal conjunctiva displayed significantly fewer myofibroblasts, but there was a distinct seasonal variation in MFB density, exhibiting a minimum during December - February and a maximum in March - May. Polyploid cells, though occasionally found in the normal conjunctiva, were significantly fewer than after allo-HSCT.

In conclusion, our results demonstrate a higher than normal MFB density in the conjunctivae of patients after allo-HSCT. The donor-derived majority of myofibroblasts increased with time post-transplant, and was found to correlate with presence and severity of ocular GvHD. Myofibroblasts are cellular mediators of normal wound-healing, but also of fibrosis and connective-tissue disease. Our data suggests that myofibroblasts may play a role in the pathogenesis of chronic ocular Graft-versus-Host Disease. The role and significance of the high amounts of polyploid cells observed during the first year after allo-HSCT remains obscure.

**Keywords:** myofibroblast, polyploid, allogeneic, stem cells, graft-versus-host disease,  $\alpha$ SMA

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