

Supplementary figure 1. Immunostaining of cross-sections of Descemet's membrane (DM) of subjects with and without FECD (Fuchs endothelial corneal dystrophy)

The expression of the top 5 of 32 proteins expressed only in the FECD-DM but not in the non-FECD-DM was validated by immunofluorescence staining of frozen DM cross-sections. The images were observed using an epifluorescence microscope with a \times 60 objective. In the non-FECD control samples, only fibrinogen α showed positive staining, whereas none of the top 5 proteins (fibrinogen α , hemoglobin α , SRPX2, tenascin-C, and hemoglobin $\gamma\delta\epsilon\beta$) were detected. Yellow arrows indicate perinuclear expression of fibrinogen α in non-FECD-DM. In the FECD-DM, fibrinogen α , hemoglobin α , SRPX2, tenascin-C, and hemoglobin $\gamma\delta\epsilon\beta$ were clearly detected by immunostaining. The nuclei were counterstained with DAPI. The right columns show the immunostaining image combined with bright-field imaging. AH indicates the aqueous humor (endothelial cell side) and ABL indicates an anterior banded layer (stromal side of DM). Immunostaining of each protein was repeated for 3 non-FECD-DMs and 3 FECD-DMs. The scale bar: 20 μ m.