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Title: Innovative strategies to clinically characterize the human tear proteome: from fundamental exploration to ophthalmological relevance
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Chapter 5: Further exploration of Proteomics for clinical tear analysis.

5.0 Introduction: Entering the Orbitrap age.

Three years after our initiation to proteomics (Chapter 4) we got access to a newer type of mass spectrometer, which we were keen to evaluate for its performance in clinical tear analysis. Meanwhile, we had been working on improvements of the tear collection procedure, making it more amenable to the ophthalmological daily practice. We selected to sample donor or patient tears no longer with glass capillaries but with Schirmer strips, which are very well accepted for clinical monitoring of DED.

As mentioned in chapter 1, Schirmer strips allow prelevation of tears with minimal irritation (even without the prior instillation of anaesthetics).

Our experiments demonstrate that only 20 μl collected on a Schirmer strip are sufficient for multiple analyses of single patient tears, thus without the need to pool samples. This is extremely useful when one intends to evaluate intra-individual changes in protein composition under different conditions, such as there are:
- technical duplicates/triplicates of the same sample;
- changes with time (diurnal, age, ...);
- effects of clinical treatments;
- correlations with patient-specific DED severity measurements (Chapter 1).

It is interesting to remark here that the bottom-up analysis data reported in Chapter 5.1 seem to further support the positive effects of our transplantation of labial salivary glands. When considering the panel of 4 biomarker proteins the absence of which can be correlated with dry eye disease as described by the well-known Singapore Eye Research Institute (Beuerman group) [Zhou et al., 2009]. Whereas none of these biomarkers were detectable in the untreated diseased state, they partly seem to get restored after the transplantation.

In addition, we want to remark here that using these modern MS systems, all our previously obtained tear protein identifications obtained by QTOF GeLC MSMS could be confirmed.

Reference