Protein Profile study of neonatal tear fluids

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Introduction

- Neonatal tear analysis can provide information about the health condition of eyes since it contains specific molecules, proteins (1).
- Protein profiles in neonates has not been studied as much as in adults tear sample due to the difficulty of collecting samples without causing reflex tearing (2).
- To explore the neonatal tear samples extensively, spectroscopic, proteomic, lipidomic, and metabolomics studies are necessary.
- Objective of the study is protein pattern analysis of term and pre-term tear fluids using high performance liquid chromatography with light emitting diode-induced fluorescence system (HPLC-LED-IF) developed in our laboratory.

Method

- Neonatal Tear Sample collection — Newborn intensive care unit, Kasturba Medical College, Manipal using Schirmer strips.
- Strip was suitably diluted in HPLC grade water (100 μL).
- HPLC Grade Solvents in mobile phase – HPLC grade water+0.1% Trifluoroacetic acid (TFA) and Acetonitrile+0.1% TFA, Requisition time-40 min, flow rate – 0.2 ml/min.
- Excitation source – 278 nm LED.
- Figure 1: HPLC-LED-IF Blockdiagram. MI-Manual Injector, OC-Optical chopper, L1, L2,L3 and L4—Lenses, CC-Control Chopper, PMT-Photomultiplier tube, MC-Monochromator, DF—Dichroic Filter, PA-Preamplifier, LA-Lockin amplifier, PC-Personal Computer.

Results

(a) Figure 2: Tear fluid protein Chromatograms of Term and Pre-Term neonates (a) and Plot of scores of factor 3 versus scores of factor 1 (b).

- The protein profiles of term and preterm tear fluid samples has been analyzed (Figure 2(a)).
- For better classification, Principal Component Analysis (PCA) was studied.
- In this method of PCA, the mean of data from all samples is first formed. The variation of each sample from this mean is calculated.
- PCA is performed with these variations. The variations from the mean for any sample are thus represented by scores for the different factors and hence scores can be used as discriminating parameter in first step.
- Discrimination between term and pre-term tear fluid samples is observed (Figure 2(b)).

Conclusions

1. Neonatal tear protein analysis using HPLC-LED-IF has been discussed.
2. The study can give the idea on tear secretion development in newborn babies by measuring the protein concentration in each sample.

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Reference