

### P05-A152 HUMAN PLATELET LYSATE FOR CORNEA ORGAN CULTURE

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**Purpose** Comprehensive concerns have been raised regarding the safety of FBS-based culture media. In this talk we discuss the benefits of using human platelet lysate (HPL) for the xeno-free culture of human donor corneas, isolated corneal stromal keratocytes (CSK) and stromal fibroblasts (SF).

**Methods** 32 human corneas unsuitable for transplantation from 16 human donors were cultured for 25-days in either 2%FBS or 2%HPL medium and compared by phase contrast microscopy (ECD and morphology), and next generation sequencing (NGS). Effects of 0.5%FBS, 5%FBS, 0.5%HPL, 2%HPL and 10%hPL on cultured human CSK and SF were evaluated.

**Results** Differential cornea culture showed lower endothelial cell loss in the 2%HPL vs. 2%FBS group (ECL hPL=-0.7% vs. FBS=-3.8%;  $p=0.01$ ). 2%HPL led to the upregulation of cytoprotective, anti-inflammatory and anti-fibrotic genes (e.g. HMOX1, SERPINE1, ANGPTL4, LEFTY2) and the downregulation of pro-inflammatory/apoptotic genes (e.g. CXCL14, SIK1B, PLK5, PPP2R3B). CSK/SF cell viability remained high in all groups (98-100%). Cell numbers and proliferation rates increased ( $p=0.024-0.001$ ), CSK marker expression decreased with higher fractions of HPL and FBS ( $p<0.001$ ). SMA1 increased with higher amounts of FBS ( $p=0.003$ ) but decreased with incremental HPL substitution in both cell types ( $p=0.014$ ). HPL contained more TGF- $\beta$ 1 (100%hPL 1.861  $\pm$  0.231ng/ml vs. 100%FBS 0.015  $\pm$  0.010ng/ml,  $p<0.001$ ). bFGF and HGF were only detectable in 100% hPL (bFGF 0.067  $\pm$  0.017ng/ml, HGF 1.074  $\pm$  0.050ng/ml).

**Conclusion** 2%HPL is a suitable xeno-free substitution for 2% FBS in human cornea organ culture, inducing less ECL and potentially beneficial alterations in gene expression. CSK and SF can be cultured with xeno-free hPL. To maintain CSK characteristics substitution must remain minimal (0.5% hPL/FBS). hPL contains the antifibrotic HGF und bFGF, suppressing myofibroblast conversion.

### P06-A143 AN ARTIFICIAL-INTELLIGENCE-BASED DECISION SUPPORT TOOL FOR THE DETECTION OF CORNEA GUTTATA ON THE DONOR CORNEAS IN THE EYE BANK

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**Purpose** Cornea guttata (CG) prevalence post keratoplasty varies from 15 to 18%, with 1 to 2% of the cases presenting with significant negative outcomes. The purpose of this research project is to create a program based on artificial intelligence (AI) that helps with the detection of CG in the donor corneas (DC) in the eye bank.

**Methods** Preoperative corneal endothelial images (PCEI) of patients who underwent keratoplasty were collected and

classified into 2 groups according to the postoperative CG grade. Group 1 included healthy corneas and those having mild postoperative CG, while group 2 included corneas with severe postoperative CG. Using previously tested semi-quantitative morphological criteria along with other characteristics such as donor age and lens status, the PCEI were analyzed and used to create and train an AI-based tool for the detection of CG. The underlying concept of the tool compares previous cases with comparable properties to the DC in test. The postoperative CG grades of previous cases similar to the DC in test determine the prediction for its CG grade. Finally, the features and CG grade of the analyzed DC are stored in the database for future use.

**Results** In total, 6221 PCEI belonging to 1078 patients were used to create a transparent and explainable decision support tool for the detection of CG through a hybrid approach combining 2 components. (1) Graphical analytic tools, whereby the PCEI pass multiple OpenCV-based image processing steps including the Watershed transform algorithm. In this step, cell membranes are delineated, and abnormally large cells or cell depleted areas are marked in red. Several other cell representations such as 'honeycomb' representation are created for an enhanced visualization of the endothelial layer (EL). (2) Machine learning (ML) classifiers including Case-Based Reasoning were created to detect CG. Initial experiments showed a performance comparable to humans (4-fold evaluation yielded precision: weighted F1 score:0.93).

**Conclusion** We presented an AI-based program able to facilitate the detection of CG in the DC in the eye bank by comparing the PCEIs with relevant previous cases, using ML classifiers and offering an enhanced visualization of the EL. The evaluation and optimization of this program will follow as the next stage of our project.

### P07-A139 GUTTAE IN CORNEAL DONOR TISSUE

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**Purpose** To report on the occurrence of guttae in corneal donor tissue.

**Material & Methods** Retrospective database study of discard reasons for corneal donor tissue at Amnitrans EyeBank Rotterdam (AER) for the period from January 2019 to December 2021 and the outcome of an eight-question survey sent to European Eye Bank Association corresponding members addressing the occurrence of corneal guttae and the practice pattern regarding donor tissue with guttae.

**Results** Between 2019 and 2021 6039 donor corneas were processed at AER. Average discard rate because of guttae in this period was 9 ( $\pm$ 4)% (n=552). Most corneas were discarded because of guttae at first evaluation (8%, n=481). Monthly discard rate because of guttae ranged from 3% to 19%. Yearly discard rates related to corneal guttae were 10 ( $\pm$ 3)%, 8 ( $\pm$ 3)% and 11 ( $\pm$ 5)% in 2019, 2020 and 2021, respectively. Average endothelial cell density (ECD) at the first evaluation from 2019-2021 was 2486 ( $\pm$ 93) cells/mm<sup>2</sup>, with average monthly ECD ranging from 2343 to 2642 cells/mm<sup>2</sup>.