

spp., which jointly comprised around 80% of the detected microorganisms. Others found in the remaining placentas were of the species: *Acinetobacter*, *Bacillus* spp., *Faklamia*, *Lactobacillus*, *Rothia*, *Micrococcus*, *Penicillium*, *Ralstonia*, *Streptococcus* and non-specific aerobic sporulating bacteria.

In contrast, 8 samples of the decontaminated cryopreserved tissue were tested positive for microorganisms with 4 placentas inhabited by *C.acnes*, 2 by *Bacillus* spp. while the remaining consisting each of the species *Staphylococcus* and *Ralstonia*.

Conclusion Overall, the decontamination measures applied during the preparation of the amniotic tissue can be regarded as effective. We found a significant reduction of the number of microorganisms detected in the amniotic tissue following antibiotic administration.

However, some of the remaining species identified in the processed samples may be considered as contamination during the preparation and testing procedures.

For instance, *C.acnes* can be considered a result of secondary contamination due to incorrect handling. Species such as *Bacillus* most likely managed to endure the decontamination process owing to its natural resilience against harsh circumstances.

P31-A150 SARS-COV-2 REAL TIME POLYMERASE CHAIN REACTION TESTING OF CORNEAS FROM POST-MORTEM SARS-COV-2 POSITIVE DONORS

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Purpose Possible transmission of SARS-CoV-2 from donors to recipients via cornea grafts is still a concern of the transplantation community. Current recommendations are to avoid corneal transplants from donors with ongoing SARS-CoV-2 infection or those recently exposed to it. During pandemic period in Croatia 21/1113; (1,9%) corneas were procured from donors positive for SARS-CoV-2 by postmortem nasopharyngeal swab tests. That tissue was discarded. Due to the lack of knowledge about the infectivity of such corneas, we started prospective study of SARS-CoV-2 presence in cornea tissue. Here we show our first results.

Methods In the study period we had four corneas procured from two post-mortem SARS-CoV-2 positive donors. For the presence of SARS-CoV-2, analysis is performed on donor serum, hypothermic storage medium and cornea tissue lysate. Corneas were stored in hypothermic condition for 8 to 10 days, after which tissue was macerated and washed with PBS. The intracellular content was released by incubation with lysis buffer, followed by centrifugation. Next, tissue lysate, serum and hypothermic storage medium were in parallel subjected to fully automated nucleic acid isolation and RNA expression was analyzed by qRT-PCR. During isolation, RNaseP was used as internal control for successful nucleic acids isolation.

Results No SARS-CoV-2 RNA was detected in the donors serum, storage medium and cornea tissue from donors who were SARS-CoV-2-positive upon tissue procurement. In nasopharyngeal swabs of post mortem positive donors cycle threshold values of viral copies were high (CT>34), indicating that

there was small number of viral particles in infected donors that could have impact on negative results in tested tissue.

Conclusion Our data suggested that corneas may not be SARS-CoV-2 permissive if the donor was postmortem positive. Further research is required to gain more coherent insight into SARS-CoV-2 transmission via corneal transplantation.

P32-A134 CYTOPROTECTIVE EFFECTS OF HUMAN PLATELET LYSATE DURING THE XENO-FREE CULTURE OF HUMAN DONOR CORNEAS

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Purpose We evaluated the suitability of 2% human platelet lysate (2%HPL) to replace 2% fetal bovine serum containing medium (2%FBS) for the xeno-free organ culture of human donor corneas.

Methods 32 human corneas unsuitable for transplantation from 16 human donors (age 69.3±15.7years) were collected 38.5±17.1 hours after death. They were first cultured in 2% FBS containing medium for 3 days (time point TP1), then evaluated by phase contrast microscopy (endothelial cell density (ECD) and cell morphology). Following an additional 25-days culture period (time point TP2) in either 2%FBS or 2% HPL medium the pairs were again compared by phase contrast microscopy (ECD and morphology), stroma and Descemet membrane/endothelium (DmE) were processed for next generation sequencing (NGS).

Results ECD did not differ between the 2%HPL and 2%FBS group at TP1 (p=0.87). At TP2 the ECD was higher in the 2%HPL group (2179±288cells/mm²) compared to 2%FBS (2113±331cells/mm²; p=0.03), and endothelial cell loss was lower (ECL hPL=-0.7% vs. FBS=-3.8%; p=0.01). There were no significant differences in cell morphology, neither between TP1 and 2 nor between 2%HPL and 2%FBS. NGS showed the differential expression of 1644 genes in endothelial and 217 genes in stromal cells. 2%HPL led to the upregulation of cytoprotective, anti-inflammatory and anti-fibrotic genes (e.g. HMOX1, SERPINE1, ANGPTL4, LEFTY2, GADD45B, PLIN2, PTX3, GFRA1/2) and the downregulation of pro-inflammatory/apoptotic genes (e.g. CXCL14, SIK1B, PLK5, PPP2R3B, SLURP1, FABP5, MAL, GATA3).

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.

P33-A110 EX VIVO MODELS OF CORNEAL EPITHELIAL REGENERATION IN BIOREACTOR: RESPECTIVE ROLES OF LIMBAL, CONJUNCTIVAL AND CORNEAL EPITHELIA

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