



bioreactor. Results: We observed that the epithelial cells (i) displayed a monolayer organization, (ii) had the appropriate polarity, tight junctions, desmosomes and microvilli, (iii) produced cytokines upon antigenic stimulation, (iv) transported nutrients and (v) differentiated into multi-lineage progeny (i.e., absorptive enterocyte, goblet and M cells). We also observed well preserved fibroblasts, lymphocytes and endothelial cells dispersed throughout the extra-cellular matrix. Conclusions: We report the development of an organotypic model resembling structurally and functionally the human intestinal mucosa. Finally, to our knowledge, previous attempts to integrate multiple cell types in constructs grown under microgravity have been unsuccessful.

#### **W.153. HIV-induced Innate Immune Responses in Uterine Epithelial Cells**

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The mucosal epithelium of female reproductive tract is the primary site of HIV transmission during heterosexual intercourse with R5-tropic strains accounting for the majority of transmission events. However, the early innate immune response of epithelial cells to HIV has not been adequately studied. Primary uterine epithelial cells were cultured on transwell inserts and stimulated apically with varying concentrations of BaL (R5) and 3B (X4) HIV for up to 48 hours. Both viruses upregulated TNF $\alpha$  mRNA expression. Interferon (IFN) beta was induced at very low levels by both viruses. Intriguingly, BaL down-regulated the production of IFN lambda while 3B had no effect. The interferon-stimulated genes (ISG) MxA, OAS2 and PKR were only induced by 3B and not BaL. This finding extended to mRNA levels of the anti-HIV molecules CCL20 and Elafin, which were induced by 3B in a dose-dependent manner. The potent innate immune response elicited by 3B (X4) in uterine epithelial cells may partially account for the increased transmissibility of R5 viruses. How BaL avoids inducing an ISG and anti-HIV response is unknown. Furthermore, the upregulation of interferon-stimulated genes in the absence of a robust IFN beta or lambda response hints at a novel response mechanism elicited by 3B.

#### **W.154. HIV Exposed Seronegative Women Express Lower Level of IFN-g Inductible Chemokine in their Cervico-vaginal Lavages**

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Over three-quarters of HIV/AIDS cases occur through heterosexual transmission. However, little is known about the factors influencing the susceptibility to HIV infection and the immune response in the female genital tract (FGT). Studies, including those with female sex worker (FSW), have reported natural resistance to HIV-1 infection. Reduced levels of immune activation, termed immune quiescence, has been associated with this resistance. The aim of this study is to analyse immune mucosal factors that could be implicated in the susceptibility to HIV infection. Methods: 213 CVL from FSW from the Majengo clinic in Nairobi, Kenya (57 new negatives (NN); 68 HIV+ and 55 highly exposed non infected (HESN)) and 33 HIV uninfected low risk women were analysed for the presence of cytokines and chemokines. Our results show a significant difference between the three groups for the chemokines CXCL-9 (MIG) and CXCL-10 (IP<sub>10</sub>). MIG and IP-10 were decreased in the CVL of the HESN compared to the other groups (ANOVA  $p=0.0002$  and  $p<0.0001$  respectively). Conclusion: These results suggest a decreased level of immune activation in the FGT of the HESN consistent with the immune quiescence hypothesis. This study will allow us to further our understanding of the mechanisms involved in the host immune response versus HIV. Funded by CIHR.

#### **W.155. Role of Toll-like Receptors in Development of Postpartum Endometritis**

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Toll-like receptors (TLRs) play a crucial role in defense against pathogens invading the female reproductive tract. Binding of specific ligands with TLRs promotes pro-inflammatory cytokine production and development of inflammation. The aim of the study was to establish the association between expression of TLR1 (ligands - triacyl lipopeptides), TLR2 (ligands - lipopeptides, lipoteichoic acid, zymosan), and TLR5 (ligand - flagellin) of cervical epithelial cells *in vivo* and development of postpartum endometritis in women with high infection risk. Materials and methods: Expression of TLR1, TLR2 and TLR5 mRNA in epithelial cells of cervix uteri was detected using qRealTime PCR on third day after delivery per vias naturalis. Samples were taken from 9 women with clinical endometritis and 17 women with normal postpartum period. mRNA was extracted using Trizol (Invitrogen). First-strand cDNA synthesis was performed using oligo dT primers and Mint reverse transcriptase kit (Eurogen). Quantitative real-time PCR was performed using qPCRmix-HS SYBR kit (Eurogen). Results were analyzed using an iCycler (Bio-rad laboratories). The threshold cycle values were normalized against the threshold value of human beta-actin and analyzed using Statistics 6.0. Results. It was shown that mean relative expression of TLR5 is significantly lower in women with postpartum endometritis compared with expression during normal postpartum period. Levels of TLR1 and TLR2 has not significant difference in both groups. Level of TLR5 was strongly correlated with data of histological results of placental tissue. Conclusions. It was shown, that decrease of TLR5 expression plays significant role in postpartum endometritis development. Research supported by grant of President of Russian Federation MK-1564.2010.7.