**Abstract for ARVO 2018**

**The interaction of uveal melanoma (UM) with Hepatic Stellate Cells (HSC).**

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**Section:** Pathology/Oncology

**Purpose:** Uveal melanoma (UM) is the most frequently occurring primary intraocular malignancy in adults. Despite successful treatment of the primary tumour, UMs have a strong propensity to metastasise to the liver, where they lead to a fatal outcome. Our previous results suggest that proteins secreted from primary UM cells played a putative role in the activation of hepatic stellate cells (HSCs). HSCs (also called Ito cells) are dormant pericytes found in the peri-sinusoidal spaces of the liver (also known as the space of Disse) during quiescent stages. Once activated, HSCs deposit extracellular matrix components, which are thought to promote tumour progression and invasion. In this study we investigated the interaction between UM cells and HSCs, and examined HSCs in metastatic UM (MUM) samples.

**Methods:** One HSC cell line, LX-2, was profiled by immunohistochemistry (IHC) to determine the expression of various markers. UM and HSC cells were co-cultured as 3D spheroids and characterised further by IHC. We also assessed α-SMA expression, a marker of HSC activation, in 23 MUM specimens by IHC. A sulforhodamine B (SRB) assay was also used to determine UM cell proliferation when challenged with 24- and 48-hour conditioned media compared to non-conditioned media obtained from a HSC cell line, LX-2.

**Results:** This study demonstrated that LX-2 cells in monolayer expressed α-SMA, vimentin, Ki67 and type IV collagen. We also showed that UM and LX-2 cells in co-culture form compact spheroids of varying size and composition depending on the UM cell line used. In MUM samples, α-SMA positivity was observed both intra-tumourally and in a peri-tumoural distribution, possibly representing the activation of HSCs by UM cells. LX-2 conditioned media increased the number of UM cells as compared with serum free medium alone.

**Conclusions:** LX-2 cells represent partially activated HSCs, which secrete factors to promote UM cell survival/proliferation. LX-2 cells can be used in co-culture with UM cells, however, further work is required to optimise the 3D-UM cell culture model to better represent the *in vivo* characteristics of MUM.

Character Limit: 2500