

## Biocompatibility of Normal Human Urothelial (NHU) and Urinary tract-derived Smooth Muscle (USM) cells grown on two and three dimensional (Poly (lactic-co-glycolic) acid (PLGA) scaffolds

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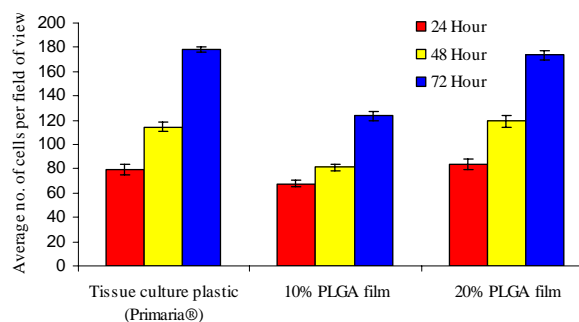
**INTRODUCTION:** The main function of the bladder as a low pressure storage reservoir for urine can be severely compromised as a result of severe dysfunction, trauma, cancer and congenital abnormalities. Tissue engineering strategies are consequently being developed in an attempt to rebuild and surgically repair the bladder. This investigation concentrates on the development of synthetic biomaterials as scaffolds for bladder repair/reconstruction. We report the preparation of non-porous two dimensional films and porous three dimensional foams and assess their biocompatibility in terms of supporting the adhesion and growth of NHU and USM cells.

**METHODS:** PLGA materials were prepared using two different methods: spin-coating to prepare non-porous 2D flat films of 10% and 20% (w/v) PLGA and emulsion freeze drying to prepare porous 3D foams. Initially, NHU and USM cells were seeded onto the 2D films and cell numbers were analysed at 24, 48 and 72 hour time points to construct growth curves. Film topography was assessed by Atomic Force Microscopy (AFM) and film degradation studies were undertaken using size exclusion chromatography. USM cells were seeded on to the 3D PLGA foams and maintained in submerged culture for up to 7 days to achieve cell infiltration and proliferation. Scanning Electron Microscopy (SEM) analysis was employed to visualise the USM cells within the 3D scaffold and USM-seeded 3D foams were embedded in polyester wax and sectioned by adapting a protocol by Steedman [1], and analysed by immunofluorescence for the expression of cell and extracellular matrix (ECM) proteins.

**RESULTS:** The 2D film study demonstrated that NHU and USM cells were able to adhere and proliferate on the PLGA non-porous 2D membranes, with both cell types growing significantly better on the 20% (w/v) PLGA films compared to the 10% (w/v) films at each time point ( $p < 0.05$ ; figure 1). AFM showed that there were no differences in surface topography between the 10%

and 20% (w/v) films. Degradation studies showed that 10% (w/v) PLGA films degraded faster due to their greater surface area to volume ratio.

*Fig. 1: NHU cell growth on 10% and 20% (w/v) PLGA 2D flat films.*



Following 7 days of incubation, SEM analysis showed that USM cells were found evenly distributed within the 3D PLGA scaffolds and immunofluorescence studies demonstrated that ECM proteins had been deposited within the scaffold in association with the cells.

**DISCUSSION:** This study has shown that PLGA is a suitable synthetic material for supporting the growth of USM and NHU cells. The 2D film study suggests that although the degradability properties can be tuned by altering the PLGA concentration, this may affect how well the materials support cell growth. We suggest that PLGA foams provide a suitable scaffold environment to investigate whether cell interactions in 3D will lead to the development of more differentiated, functional tissue facsimiles. However, methods to monitor and assess the growth and phenotype of cells seeded in 3D scaffolds will require further development.

**REFERENCES:** <sup>1</sup> HF.Steedman (1957) *Nature*

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