Comparison of two cell culture set-up for identification of optimal textile scaffold regarding cell response

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INTRODUCTION: Scaffolds are designed to be temporary structure providing cells a framework and guiding structure. They are used for *in vitro* and *in vivo* tissue engineering [1]. Textile based scaffolds have the advantage of having defined porosity and structural elements (fibres). So far it is still unclear how the scaffold structure influence cell behaviour. The few existing reports mention that at least the fibre diameter is able to greatly affect cell migration and cell shape [2, 3].The aim of the present study is to identify optimal textile scaffold characteristics regarding the biological response. For this we investigated the influence *in vitro* of surface chemistry, fibre diameter and interfibre mesh spaces of woven textile scaffolds on the behaviour of primary adult human osteoblasts and dermal fibroblasts. The biological effects were characterised using a common test-set-up measuring total number and percentage of proliferating cells seeded as single cells on the scaffold. Furthermore, a new test set-up was developed mimicking the *in vivo* situation more appropriate, i.e. by placing cell reaggregates onto the scaffolds and assessing the areacell outgrowth.

METHODS: *Samples:* Five types of plasma cleaned woven fabrics were used for this study. Two fabrics were made of polyethylene terephthalat (PET) and three of polyamide 6.6 PA (Sefar, CH). The fabrics varied regarding fibres diameters (42-45 versed 77-86µm) and distance between fibres (100-105 versed 200µm). *Cell culture:* Primary normal human dermal fibroblasts (NHDF) were purchased from Cambrex and primary adult human bone cells (HBC) were obtained by cultivating trabecular bone pieces from patients receiving hip prosthesis. *Cell proliferation:* Single cells were seeded onto the nets. The culture flasks were kept for 24 hours on the gyratory shaker to obtain a homogeneous cell distribution. In the following 7 days cells were cultured under static conditions. The proliferating cells were labelled by adding BrdU 24 hours before immunostaining on day 1, 4 and 7. The cells were additionally stained with DAPI to visualize the nuclei of all cells. The BrdU positive and negative cells were counted using a fluorescent microscope. *Cell spreading:* Cell reaggregates were prepared by gyratory shaking of cell suspensions for three days under cell culture conditions. Single reaggregates were seeded onto the scaffolds and kept under cell culture conditions for 10 days. Cell outgrowth was examined every day. The area was measured using Motic Images Plus 2.0 software. The cell outgrowth was expressed as a percentage of the projected area of the reaggregate on the first day of culture.

RESULTS & DISCUSSION: We could show that HBC and NHDF could attach, proliferate on the PET and PA fabrics with various diameters and distance between fibres. In comparison to the common test set-up, the new test set-up (measuring the cell outgrowth area starting from a cell reaggregate seeded on the scaffold) was detecting differences in scaffold characteristics with greater sensitivity. Only with this new set-up we were able to distinguish between the biological effects of two different fibre diameters (42-45 with 77µm) and surface chemistries (PET and PA), with the PET fabric with 42 fibre diameter being of the evaluated materials significantly the best scaffold in promoting cell performance.

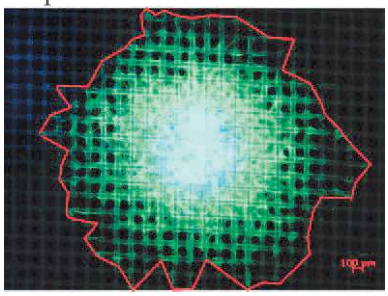

Fig. 1: Fluorescence images of reaggregate cultured of NHDF on net fabrics PET 42/105 after 7 days in culture (green: F-Actin, blue: DAPI stained). Red line: the outgrowth area

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