VERIFICATION WHETHER THE POROUS STRUCTURE MANUFACTURE WITH AM METHOD CAN BE SUITABLE FOR CELL CULLTURE

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Introduction

The use of 3D additive manufacturing (AM) technology for the production of lab-on-a-chip systems is not a widespread technology [1] and is still under development. A growing number of publications has demonstrated the usefulness of 3D printing as a method for producing micro bioreactors [2, 3] Currently, commercial 3D printing devices already offer printing resolution of 1-200 µm, and may use biocompatible materials.

Methods

We subjected a SLS printout to topographic and biological tests. Contrary to solid and liquid base methods, SLS does not require designing a porous structure in the CAD model [7], but it allows for obtaining a porous structure by properly setting the process parameters, that is the powder fragmentation. The designed 3D geometry is presented in Figure 1.

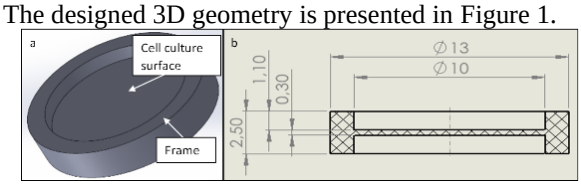

Figure 1: Culture disc: a) 3D model of a structure suggested for use in tissue engineering, b) dimensions of the culture disc

The PA2200 powder we used had a PSD of approx. 56 µm and is biocompatibility material. We used the WEHI 164 mouse Cell Line obtained from fibrosarcoma after 32 passages. Five culture discs were subjected to biocompatibility analysis. They were placed in wells A2 to A6, and the A1 well was designated as the reference well. We cultured cell in all wells. After 48 h and 96 h, the cell culture medium in the wells was replaced and microscopic observations were conducted after a week of the incubation.

Results

Based on our assumptions and previous results, the SLS increment technology allowed us to obtain the structure characterized by spatial and surface porosity which turned out to be suitable for cell culture of WEHI 164. The obtained disc structure was characterized by roughness (Sa) of 11.65 µm, a pore size of up to 100 µm, and an average pore size of 37.5 µm (Figure 2). The analysis was carried out on images captured at a 500× magnification using the software included with the Keyence VHX 750 microscope. Due to the preliminary nature of the research and the need to first determine the material properties and the process parameters, the assessment of whether the printed structure promotes the development of cells was only qualitative and based on the comparison of the culture medium colour in the course of the experiment. The colour of the culture medium changed evenly

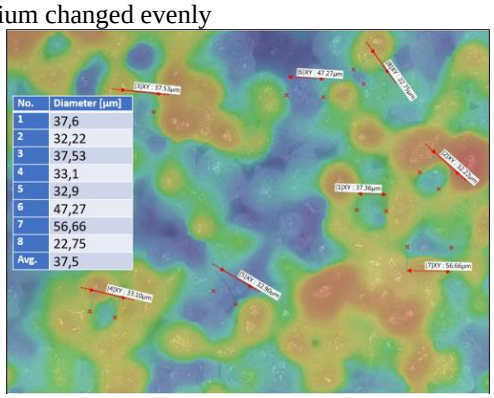

Figure 2: Analysis of the surface structure with marked size of sample pores.

It has not been possible, to detach all cells from the discs without destroying the material or damaging the cells, which may further indicate the positive effect of the structure on cell adhesion. Thus a viability test was performed by staining the cells.

Discussion

The SLS technology allowed us to achieve the desired porosity. Our previous experiences suggested a need for designing the disc with a frame; the idea was to help to ease cell culture regardless of the used culture vessel type To separate the cells from the structure, we performed trypsinization. We concluded that trypsinization did not reach all the pores of the structure, probably because the porous structure prevented the cells from leaving certain spaces during the incubation. The AM, which involves sintering PA2200 powder with a laser beam, offers new possibilities for producing convenient surfaces for tissue engineering. Based on micro- and macroscopic examination and observations, it can be concluded that the use of this additive printing technology and this specific printing material offers a wide range of possibilities for future applications in tissue engineering, both in the context of cell culture surfaces and culture platforms.

References

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