Purpose of the visit

Purpose was to practice my skills to do research abroad and to work with international research groups and also meeting and discussing with other researchers in same research which would broaden my knowledge on this area and thus complement my PhD student skills. The visit idea was also strengthen the collaboration with University of Erlangen-Nuremberg and University of Helsinki and increases my scientific network. The main scientific work was cellular and bacterial footprint analysis on different biomaterials (titanium, diamond like carbon and diamond like carbon polytetraflouroethylene hybrid) with time of flight secondary ion mass spectroscopy (ToF-SIMS) equipment. Results from these experiments is planned to be included to my last thesis publication.

Description of the work carried out during the visit

ToF-SIMS measurements were done to all samples for all planned time points: Titanium, DLC and DLC-PDMS-h, time points 0, 1, 7, 14, 21 and 28. 30 samples altogether. Positive and negative static SIMS measurements were performed using a ToF-SIMS spectrometer (ION-TOF, Münster) on all sample surfaces.

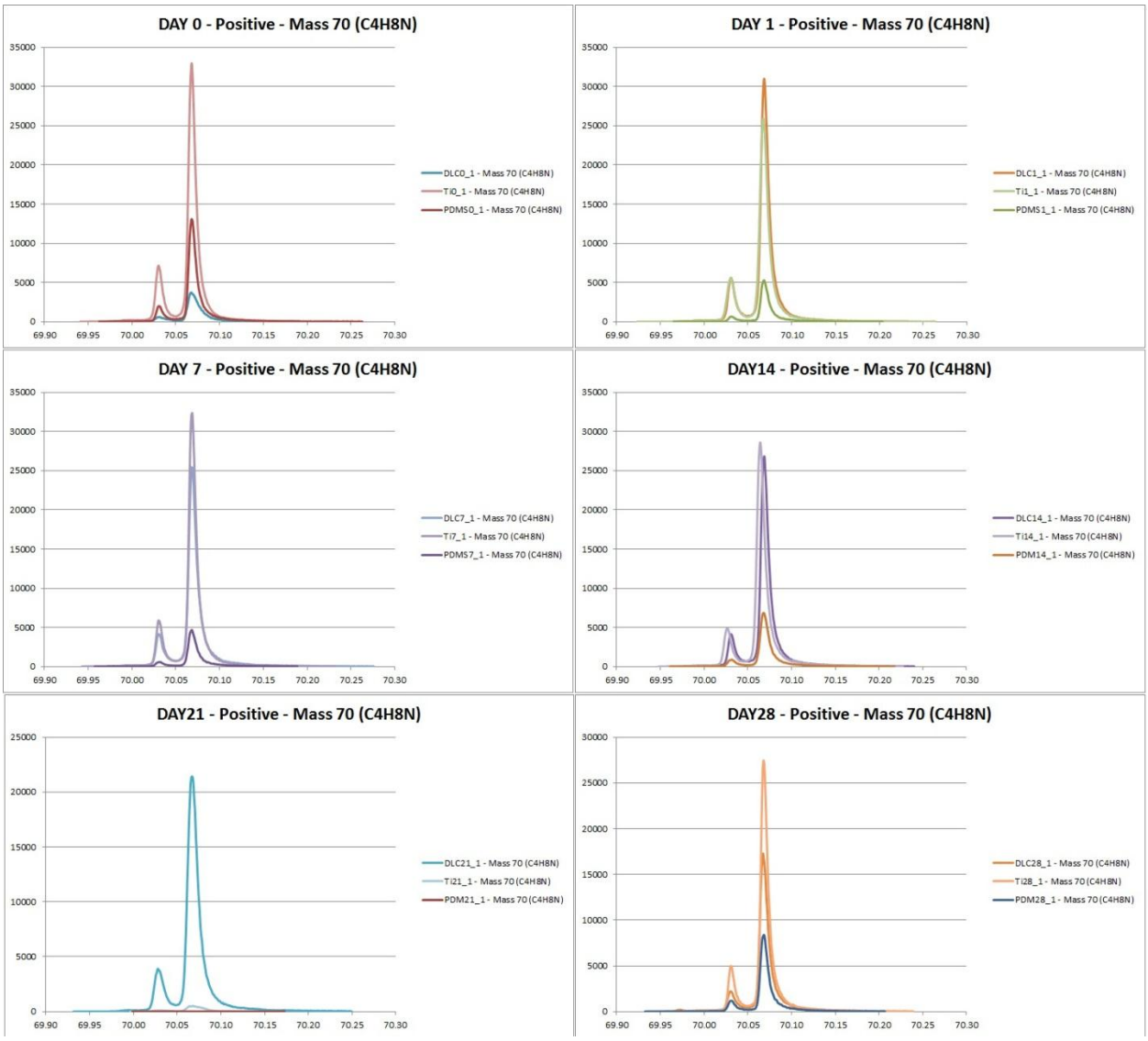
1) The samples were irradiated with a pulsed 25 keV Bi^{3+} liquid-metal ion beam. The beam was electrodynamically bunched down to 25 ns to increase the mass resolution and rastered over a 500 x 500 μm^2 area.

2) measurements were made separately from pattern and background, from an area of approx. 70 x 70 μm^2. The primary ion dose density (PIDD) was kept at $\sim 5 \times 10^{11}$ ions x cm^{-2}, ensuring static conditions. Signals were identified using the accurate mass as well as their isotopic pattern.

Description of the main results obtained

We have earlier hypothesized that adhering microbes would leave biomolecular footprints on implant surfaces. We had developed a novel diamond like carbon polydimethylsiloxane hybrid (DLC-PDMS-h) coating. In studying its cytocompatibility using osteoblast Saos-2 cells we noticed that Saos-2 cells did not spread on DLC-PDMS-h substrate and after a few days of culture Saos-2 cells underwent apoptosis , starting to express activated effector caspase-3 and undergoing blebbing, formation of apoptotic bodies, nuclear condensation (pycnosis) and fragmentation (karyorrhexis), followed in cell culture by secondary necrosis. In contrast, osteoblast precursors, mesenchymal stromal (stem) cells MSCs did according to scanning electron microscopy adhere to DLC-PDMS-h and although their spreading did not develop as well as on e.g. titanium or DLC, they did not undergo apoptosis.

Based on the above we had developed a hypothesis, which suggested that DLC-PDMS-h substrate does not allow the mesenchymal Saos-2 cells to develop focal adhesions to adhere to its substrate. Mesenchymal cells need such cell-extracellular matrix (ECM) contact and if they are not able to develop such cell-ECM contact they undergo a special form of apoptosis, known as anoikis. Obtained results strongly suggest that our hypothesis is correct. Consistently all different mass points did show lower amount of mass on DLC-PDMS-h material surface than on other tested biomaterials, see e.g. mass 70 (figure below).



Future collaboration with host institution (if applicable)

Future ToF-SIMS measurements are being planned and other collaboration possibilities are under consideration.

Projected publications / articles resulting or to result from the grant (ESF must be acknowledged in publications resulting from the grantee's work in relation with the grant);

The results will be included in my last and final thesis publication which is in manuscript phase. Current title: "Osteogenesis on DLC-PDMS-h surface". Naturally ESF will be included in Acknowledgements.