

Influence of topography and wettability on biocompatibility

This application note illustrates how the Attension Theta Optical Tensiometer combined with the 3D topography module can be utilized to study biomaterials.

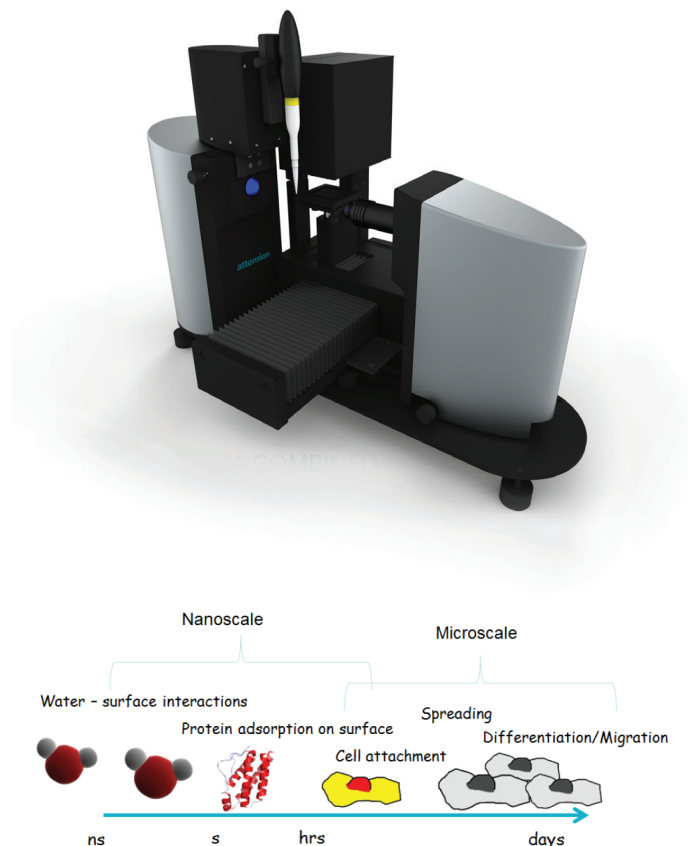
Introduction

Various types of artificial materials are being utilized as implants in all fields of medicine. The surface properties of the implant determine its interactions with the surrounding host tissue. Physicochemical properties of the surface, like wettability and surface topography, are of prime importance for the optimization of adhesion, spreading and proliferation of cells. Different types of metal substrates like stainless steel, titanium and titanium alloys have been utilized as implant materials, especially for dental and bone implants. Polymers are often used either together with metals in hard tissue replacements or in applications where mechanical durability is not the needed [1].

The initial response when material is placed in the biological surroundings is the water molecule adsorption to its surface. This happens within the first few nanoseconds. In the second stage the protein adsorption occurs. It is generally accepted that the small proteins will be the first to adsorb due to their rapid transport to the surface. Over time these proteins are replaced by bigger ones that have greater affinity towards the surface [2]. Wettability of the substrate is known to influence protein adsorption. It is usually reported that biomaterial surface with moderate hydrophilicity improved cell growth and higher biocompatibility. However, cell adhesion can decrease as the material becomes very hydrophilic. This points out the existence of a range of optimal surface energies [3]. The third stage of biological response includes the cell attachment to the surface. This stage is influenced by adsorbed protein layer as well as surface topography. Cell spreading and differentiation is known to be influenced especially by both micro scale roughness and wettability. It is thus important to be able to determine when the cell spreading is modulated by surface free energy, topography or both [4]. A summary of the host –biomaterial interactions is presented in Figure 1.

Since surface roughness is known to play a role in cell – biomaterial interactions, lots of effort has been put into determining the most suitable roughness parameters for implant

surface characterization. Wennerberg and Albrektsson [5] proposed the use of 2D height parameters, R_a and R_q and their 3D counterparts S_a and S_q , respectively. For hybrid parameters the use of S_{dr} was proposed.



[Fig. 1]: Implant - host interactions dependence on surface roughness scale.

Case study 1: Staphylococcal biofilm growth on smooth and porous titanium coatings

Titanium is a commonly used metal in bone implant applications. For osseointegration it is an advantage that the surface of the implant is porous. It allows the bone cells to grow in the pores supporting an improved anchorage with the surrounding bone tissue. It is however also well known that porosity will increase the surface roughness, which is associated with an enlarged risk on bacterial adhesion. In the study by Braem et al. [6], the roughness, wettability and porosity of titanium coatings were studied for decreased Staphylococcal biofilm growth.

In this study nine different titanium surface coatings were studied. State-of-the-art porous VPS Ti was used as a reference. Other coatings were produced by using TiH₂ starting powders with different grain size; Vm = 10 µm and P = 40 µm. The surface roughness of the coatings varied from very smooth (S_a = 0.03 µm) to extremely rough (S_a = 27.83 µm). Contact angles were measured with the CAM 200 (Attension, Biolin Scientific) and show values from completely wetting to 143°. Live cells were counted on each titanium surface and compared to the reference surface (VPS Ti). Surface roughnesses, contact angles and the live staphylococcus aureus count after 72 hours of incubation are presented in table 1.

The results indicate that the average surface roughness, S_a, and hydrophobicity (measured by water contact angle) are predominant factors positively affecting the biofilm growth. From table 1 it can be seen that the surfaces EPD Ti (P), EPD Ti (Vm/Vm) and EPD Ti (P/Vm) exhibit relatively large bacterial colonies on their surface. This is both because of the high surface roughness as well as the hydrophobicity of the surface. The best results are obtained with the rough titanium surface coated with hydrophilic coating. This surface combines the favorable surface texture to hydrophilic chemistry making it a possible candidate as a bone implant surface.

[Table 1]: Surface roughness, contact angle and live cell count for selected titanium substrates.

Sample	S (µm)	S (%)	Contact angle	Live cells (72hr)
VPS Ti	27.8	720	143	1
Polished Ti	0.03	0	47	0.07
Beadblasted Ti-641-4V	5.8	53	46	0.08
EPD Ti (Vm)	1.6	27	92	0.06
EPD Ti (P)	4.5	62	102	0.29
EPD Ti (Vm/Vm)	6.9	106	109	0.24
EPD Ti (P/Vm)	8.0	219	106	0.88
EPD Ti (P)* MAO	4.3	36	-0	0.05

Case study 2: Utilization of 3D topography module in biomaterial research

When contact angles are measured on a rough surface, a correction should be done according to Wenzel equation [7] to be able to separate the wettability caused by the surface roughness from the chemistry. 3D topography combined with Theta optical tensiometer provides a unique tool for the combined surface roughness contact angle measurements. In table 2 S_{dr}, measured contact angle and corrected contact angle from four different titanium surfaces are presented. When surface roughness measurement is done in conjunction with the contact angle measurement, it is possible to do both measurement on exactly the same sample spot. The corrected contact angle is calculated automatically by the software.

[Table 2]: S_{dr}, measured and corrected contact angle presented for four titanium surfaces.

Sample	S _{dr}	Measured contact angle	Corrected contact angle
Ti 1	22	96	95
Ti 2	41	107	102
Ti 3	65	103	98
Ti 4	78	110	101

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