dataphysics

DataPhysics Instruments GmbH · Raiffeisenstraße 34 · 70794 Filderstadt, Germany
phone +49 (0)711 770556-0 · fax +49 (0)711-770556-99
sales@dataphysics.de · www.dataphysics.de

Application Note 24

Protein-Adsorption on Surfaces

Problem

In many areas manmade surfaces come into contact with biological systems. This can be for example the hull of ships in seawater, machinery and packing materials in the food production or implants into tissue. All of these surfaces will be covered by a layer of biological components in a short time.
Proteins do have an important influence to the creation of this initial bio layer, as they are the first to bind with the surface. Afterwards this layer of proteins will be a good connection for other microorganisms to settle on the surface and may cause damage to it.
The protein adsorption is therefore an important interest for various areas of application like the food production, biology, biotechnology or medicine.

Method

With the DataPhysics Tensiometers of the DCAT-series it is possible to observe the adsorption behavior of proteins to the surface. Also it is possible to do studies of reversibility afterwards. This gets available by the continuous measurement of dynamic contact angles with the Wilhelmy-Method.
Due to the increasing amount of proteins at the surface the surface characteristics will change and therefore the measured contact angles as well. For this kind of measurement the absolute values of advancing and receding contact angles are not important, the information will come through the hysteresis between them.

$$H = \theta adv - \theta rec$$

Over time more and more proteins will bind to the surface. At some point there will be an equilibrium between adsorption and desorption, so as many proteins bind to the surface as many will unbind as well. As the protein coverage will be constant from this point on, the characteristics of the surface will also stay the same. Therefore the measured contact angles and the hysteresis between them will be unchanged, as seen in figure 1.

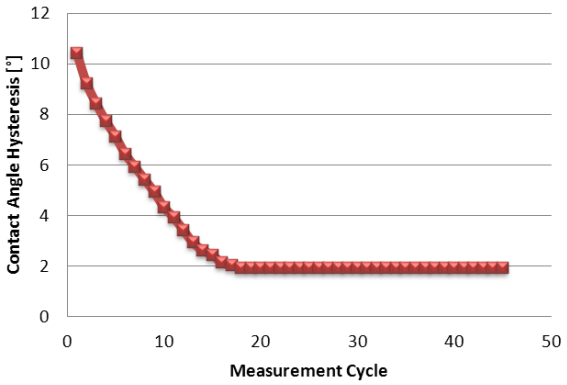

Figure 1: Example of the Contact Angle Hysteresis during protein adsorption

The measurement will be divided in different steps:

1. At the beginning of the measurement some cycles in clean/protein free liquid will be measured. These measurements will be used as a reference for the clean surface for comparisons at the end.
2. The main step is the measurement of several cycles in the protein solution of interest. Depending on the used surface and proteins it can take some time until equilibrium of adsorption and desorption will appear. Therefore it can be necessary to do a high number of measurement cycles (up to several hours).

The studies will end after some more measurements with clean/protein free surrounding liquid. The reason for this measurements is to study the desorption behavior of proteins from the surface. This third

step will be repeated several times with new protein free liquid until there is no change in the measured values anymore.
The advantage of this method is the free adsorption and desorption of the proteins to/from the surface without any external force applied on them.

Result

As an example figure 2 shows the measurement graph for the observation of protein adsorption and desorption to/from a surface. As mentioned before the measurement was divided in three steps. First of all there have been done some measurements with protein free liquid to get the knowledge about the values of the clean surface; this is shown with the measurement cycles -10 to -1.
In the adsorption phase (until cycle 45) there will be an adsorption of the proteins to the surface. It can be seen that the hysteresis stays constant after cycle 18. From this point there is an equilibrium between adsorption and desorption to/from the surface.
After the adsorption phase there have been done additional cycles in protein free solution. During these cycles the proteins attached to the surface get the chance to unbind again. To offer protein free liquid over time the liquid will be exchanged every five cycles. In the figure 2 it is shown that the contact angle hysteresis will not change anymore after the third desorption phase. The free desorption of the protein came to an end at this point. If you compare the hysteresis values recorded at the beginning of the measurement (cycles -10 to -1) with the values at the end of desorption it can be seen that the values (and therefore the surface) it not identical to the beginning. The protein adsorption is not completely reversible and some proteins will stay at the surface.

Conclusion

With the DataPhysics tensiometers of the DCAT-series it was possible to observe the adsorption and desorption behavior of protein to/from a surface. This method offers the possibility to get information about the process of the creation of biological layers at any surface. Due to surface treatments, coatings etc. it is possible to change these processes or even exclude them completely. Information that can be gathered by this method are e.g.:

- Adsorption kinetics (when/how fast is equilibrium of adsorption and desorption reached?)
- Reversibility (will the protein layer unbind completely?)

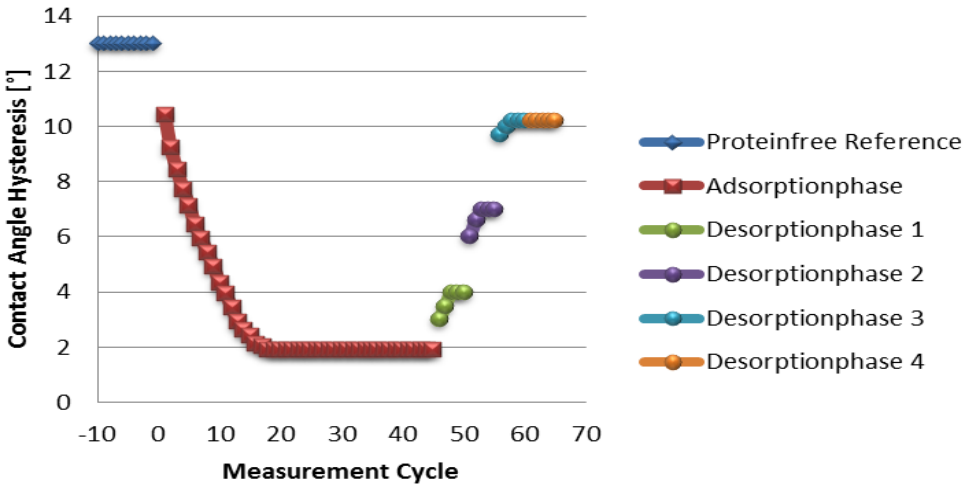

Figure 2: Graph of a complete Adsorption-/Desorption- measurements