Considerations
on
Noise and Baseline drift

Surface plasmon resonance is an optical technique that measures, refractive index (RI) changes on the interface of a metal surface, like silver, copper, gold or aluminium, and a solution. Several sources influence noise and baseline drift of the SPR signal. The instrument itself, the electrical circuits and optical path, has it own noise and drift, but those have been minimized. More severe sources are from experimental conditions, like air bubbles, temperature fluctuations, flow rate changes, sample injection, contamination or sensor design. In this application note, some of those sources will be addressed, and possible solutions will be given to minimize noise and drift.

Cleaning the sensor surface

Not only used, but also new gold coated sensor disks need to be cleaned before they can be used in an experiment. Not cleaning the disk may lead to poor SPR signals or high signal drift and noise. A few possible cleaning procedures are discussed below.

Piranha Acid

Incubation of the bare gold disk in Piranha solution, containing sulphuric acid and hydrogen peroxide in a 70% : 30% vol ratio, for 2 minutes. The disk needs to be rinsed and/or sonicated with double distilled water and thoroughly dried with nitrogen after that. This procedure will make sensor disk hydrophilic.

Base / detergent mixture

Sonication of the gold disks in a base/detergent solution, 0.1M NaOH and 1%Triton X-100, will gently clean the disk in 10 minutes. It is also possible to perform this procedure while the disk is mounted in the instrument, by using a wash sequence (wash needles and cuvette.seq).

Alcohol

Putting the disk in alcohol (ethanol or propanol) for 30 minutes and then in hexane for 2 minutes, will remove all organic contaminants.

Electrochemical pre-treatment

Cleaning the sensor disk electrochemically is possible by using the electrochemical cuvette in the instrument. First the disk needs to be cleaned with ethanol and water and then electrochemically by cycling 500 times between –0.6V to 0.0V (vs. solid Ag/AgCl) at a scan rate of 5.6 V/s. Before every measurement, a pre-treatment at –0.60V for 180s can be done for optimised performance. Please note that electrochemical treatment of the disk at high potentials (> 0.5 V) may strip Au from the surface and decrease the quality of the SPR signal.

Plasma cleaning

This is a two-step procedure: (i) exposure of the Au surface to UV light/ozone (or O_2 plasma) and (ii) immersion in pure ethanol (6,7). Organic contaminants present on the gold surface will be oxidized in the first step. In the second step the gold oxide, formed on the Au surface during the UV/ozone treatment, will be reduced to Au. The UV/plasma treatment (3,4,5) can also be used to remove organic thin films from the gold surface, including surface bound proteins from previous experiments.

All the cleaning procedures described above may be used stand alone or in combination. Electrochemical pre-treatment (1,2) or plasma cleaning (3,4,5,6,7) can also be used to remove a thiol layer. The other procedures could also be used for removing bio-molecules.

Temperature Fluctuations

Since RI is sensitive to temperature, increase of temperature will decrease the signal and cause the SPR signal to drift. The ESPRIT and SPRINGLE systems have an optional temperature control, although most measurements are usually performed at room temperature. By using the reference channel in the ESPRIT system, the data can be corrected for systematic errors. It is important to bring all buffers and samples to operation temperature before running an experiment.

The instrument should not be operated adjacent to air conditioning and heating vents.
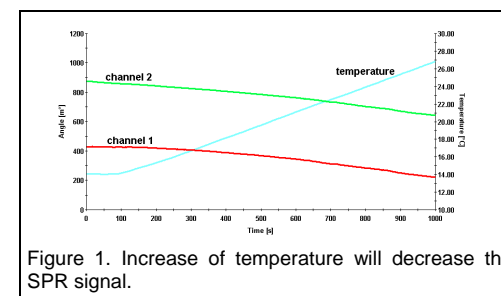

Figure 1. Increase of temperature will decrease the SPR signal.

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Sample injection

The first seconds after injection of a new sample, a decay in the signal can be observed. This is caused by a temperature difference between the cuvette and the sample. This can be prevented using thermostated needles, which is a new addition to the ESPRIT system.
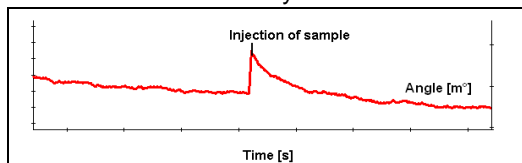

Figure 2. Angle shift due to injection of sample without temperature control.

Flow rate change

A change in mix rate will cause an immediate shift in the signal. It is therefore important to have the same flow rate throughout the whole experiment.
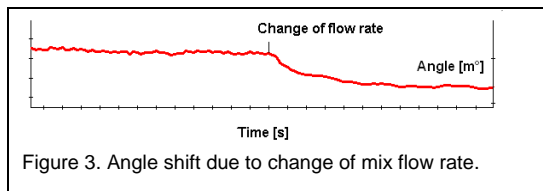

Figure 3. Angle shift due to change of mix flow rate.

Buffer and air bubbles

A change in buffer will change the RI and therefore the SPR signal. Long measurements in an open cuvette will cause evaporation of the solvent and thus increase in concentration; this can and should be prevented. Close open buffer flasks and decrease the opening of open vials before running experiments on a large number of samples.

Hydrophobic layers can cause air bubbles to exist after injection of samples. An air bubble will prohibit good liquid contact with the surface and will thus negatively influence the quality of the SPR signal.

Matrix effects

Thiol layers, dextran layers and surfaces with immobilized ligands all need stabilization procedures to minimize matrix effects. Running an experiment, the layer will go through changes in pH or ionic strength (high-low salt concentrations). These changes will influence the SPR signal. Exposing the layer to the different buffers used in the experiment for a period of time before the actual interaction, will lead to more stable signals.

General remarks

Use of deionised or purified water to prepare buffers is recommended. Filtered liquids, buffers, and samples help to prevent suspended matter from randomly attaching to the surface and influencing the SPR signal. Degassing the solutions will also contribute to more stable results.

Literature

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