Prevention of Non-specific binding as a Way to Increase Sensitivity of SPR-based Biosensors

S.B.Shankar Ganesh[1], **Valery A. Petrenko**[2]
&
Aleksandr L.Simonian[1]

[1]Samuel Ginn College of engineering
Materials engineering department
[2]College of Veterinary Medicine
Department of Pathobiology
Auburn University, AL 36849

The elimination of background signal arises from non-specific binding is a pre-requisite for the development of affinity-based biosensor for field applications as this determines the credibility of the sensor signal. Several different compounds like bovine serum albumin (BSA) and Casein were suggested to reduce non-target binding on various surfaces, such as gold, glass, silicon, etc. However, because BSA and Casein contains IgG that may become an antigen for cross-reacting secondary antibodies, they are not able to provide complete solution to the problem. Poly (ethylene) glycol has proven to be an excellent candidate for significant reduction of non-specific adsorption. Here, we investigated the properties of comb like structure of PLL-g-PEG to improve prevention of non-specific binding on the gold surface of SPR sensor, thereby reducing the background signal and improving the sensitivity of detection. We modified presented earlier method of PEG synthesis with Poly-L-Lysine (which is positively charged under physiological pH 7.4), to create a comb-like structure of PLL-g-PEG. This cationic polymer was grafted over anionic self-assembled monolayer formed by 11-MUA to prevent adsorption of unwanted species to the sensor surface. *Staphylococcus aureus* was selected as target bacteria because of its environmental importance (pathogenicity) and for its size (~1µm). Surface plasmon resonance sensor (SPREETA^{TM}) was used to study the adsorption kinetics of this bacteria to gold surface modified with MUA - PLL-g-PEG layers. Reduction in bacterial adsorption on sensor was comparable with observed reduction for the surface modified with BSA.

PLL-g-PEG was then realized as non-specific blocker in the investigations of binding kinetics in affinity interaction of β-galactosidase with specially selected phage. The dissociation constant obtained with SPR sensor modified with physisorbed PLL-g-PEG (K_d = 26.8 nM) agree very well with values reported in the literature (K_d = 30nM in ELISA). Significant increase in sensitivity by three folds (up to pico-molar) was observed compared to traditional blockers (BSA-Casein – up to nano-molar). Hence, PEG modified with PLL shows large potential application in the field of biosensor in the characterization of "guest-host" interactions.

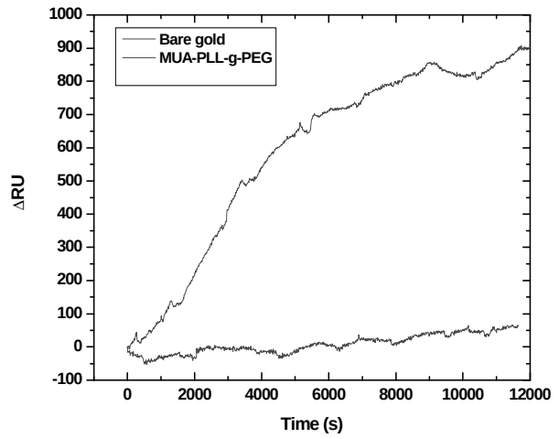

Figure 1 Reduction of bacterial adsorption (1X10[1] cfu/mL to 1X10[9] cfu/mL) on gold through PLL-g-PEG modification

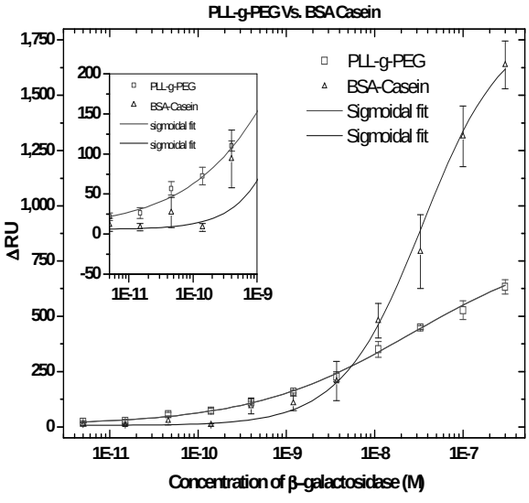

Figure 2. Traditional BSA-Casein blocker Vs PLL-g-PEG Polymer

Acknowledgement:
Supported by AUDFS Center of AU Peak of Excellence (A.S)