

Design of advanced technologies of receptors immobilization on the gold electrode surface

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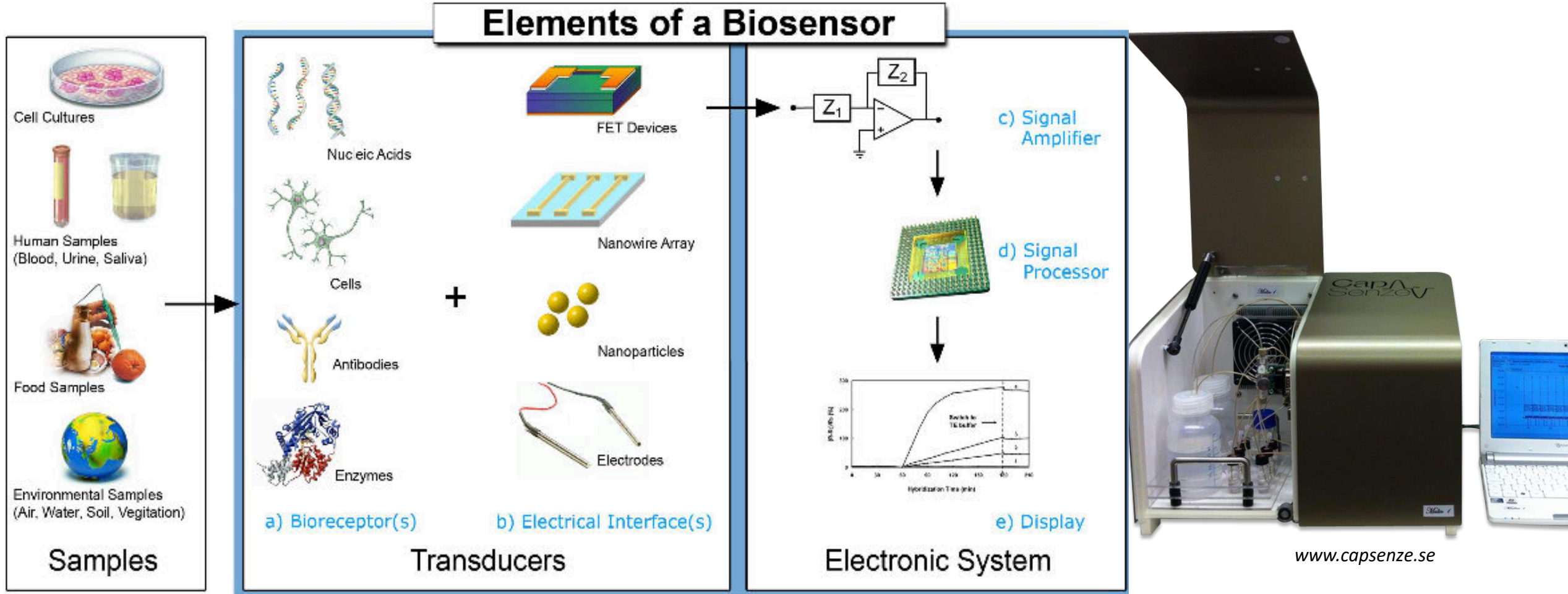
14-16 Laboratory of Biosensors and Bioinstruments, Nazarbayev University (Astana, Kazakhstan)

18-19 Laboratory of Biosensors Department of microbiology Division of Bioscience, Aarhus University, Denmark

MINES ParisTech, PSL Research University, Center for Materials Forming (CEMEF), Sophia Antipolis, France 2022

INTRODUCTION

BIOSENSOR is an analytical device, used for the detection of an analyte, that combines a biological component with a physicochemical detector.



Comparison of various types of biosensors by sensitivity

Analytical methods used for monitoring impurities in protein pharmaceuticals

Analytical techniques	Linear dynamic range (ng/ml)	Limit of detection (ng/ml)
Capacitive biosensor	0.001–1	0.001 (fg/ml)
SPR	100–10 000	20
QCM	50–1000	16
Flow-injection ELISA	0–300 000	not reported

Concept of signal detection by Capacitive Biosensor

$$\frac{1}{C_{final}} = \frac{1}{C_{double\ layer}} + \frac{1}{C_{insulated\ layer}} + \frac{1}{C_{receptor}} + \frac{1}{C_{analyte}} + \frac{1}{C_{nonspecific\ ads}}$$

Equation 1. Calculation of total capacitance

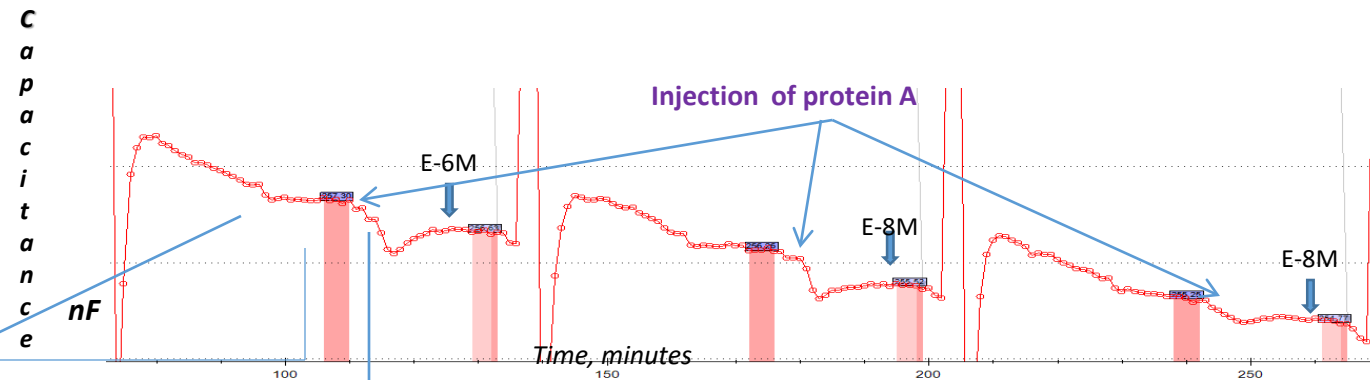
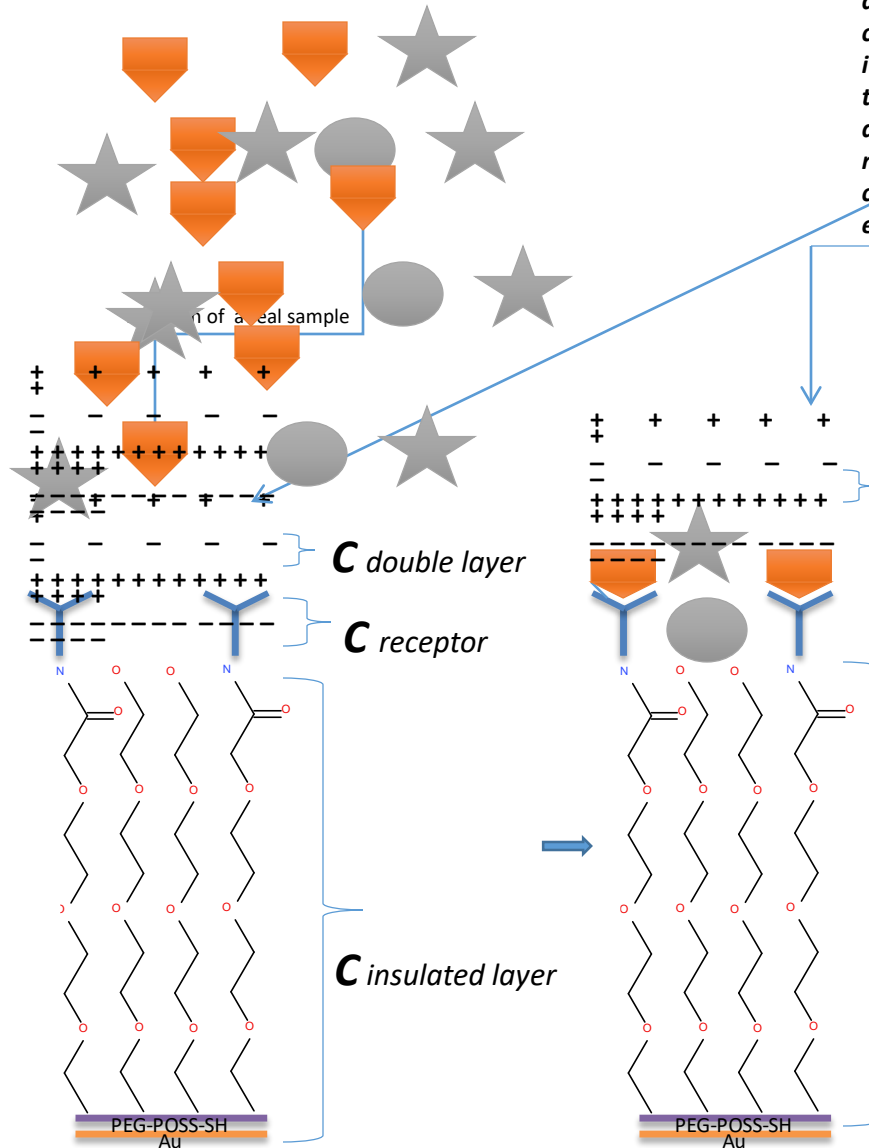


Figure 1. Response of capacitance for electrode with immobilized antibodies (gold surface covered by PEG (500 Da), on injections of regeneration buffer (25mM glycine solution, pH2.5) and given concentration of analyte.

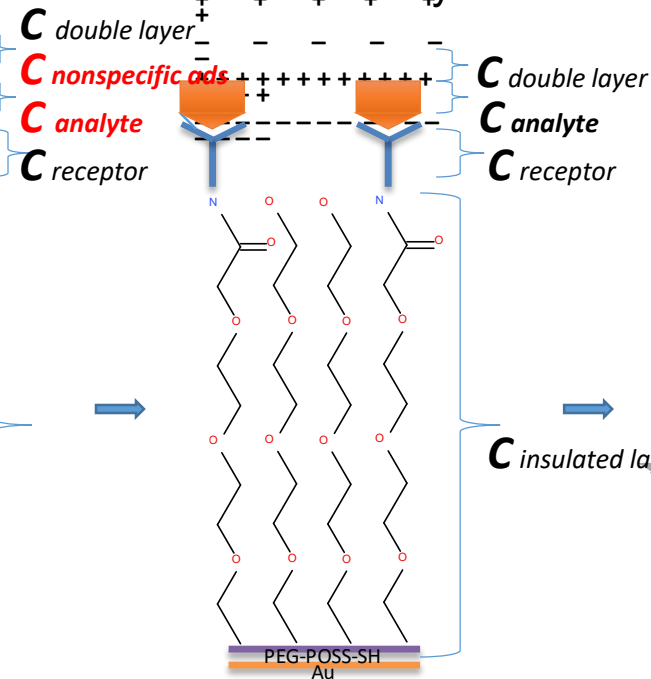


Figure 2. Concept of signal detection by Capacitive Biosensor

GLUCOSE DETECTION as concept of detection of small molecules

Figure 3. Glucose injection (10^{-15} M) into the immobilized APBA capacitive electrode under optimum conditions (Flow rate $100 \mu\text{L min}^{-1}$; **sample volume 250 μL** ; running buffer 10 mM phosphate, pH 8.5; T 25 °C).

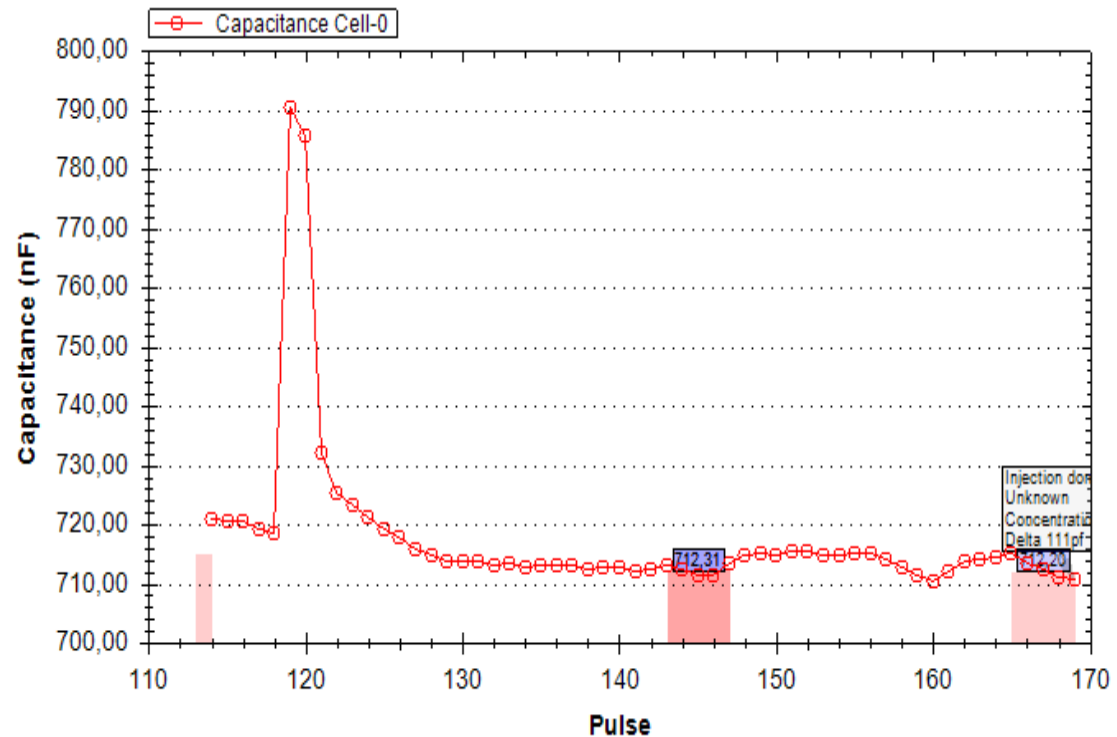
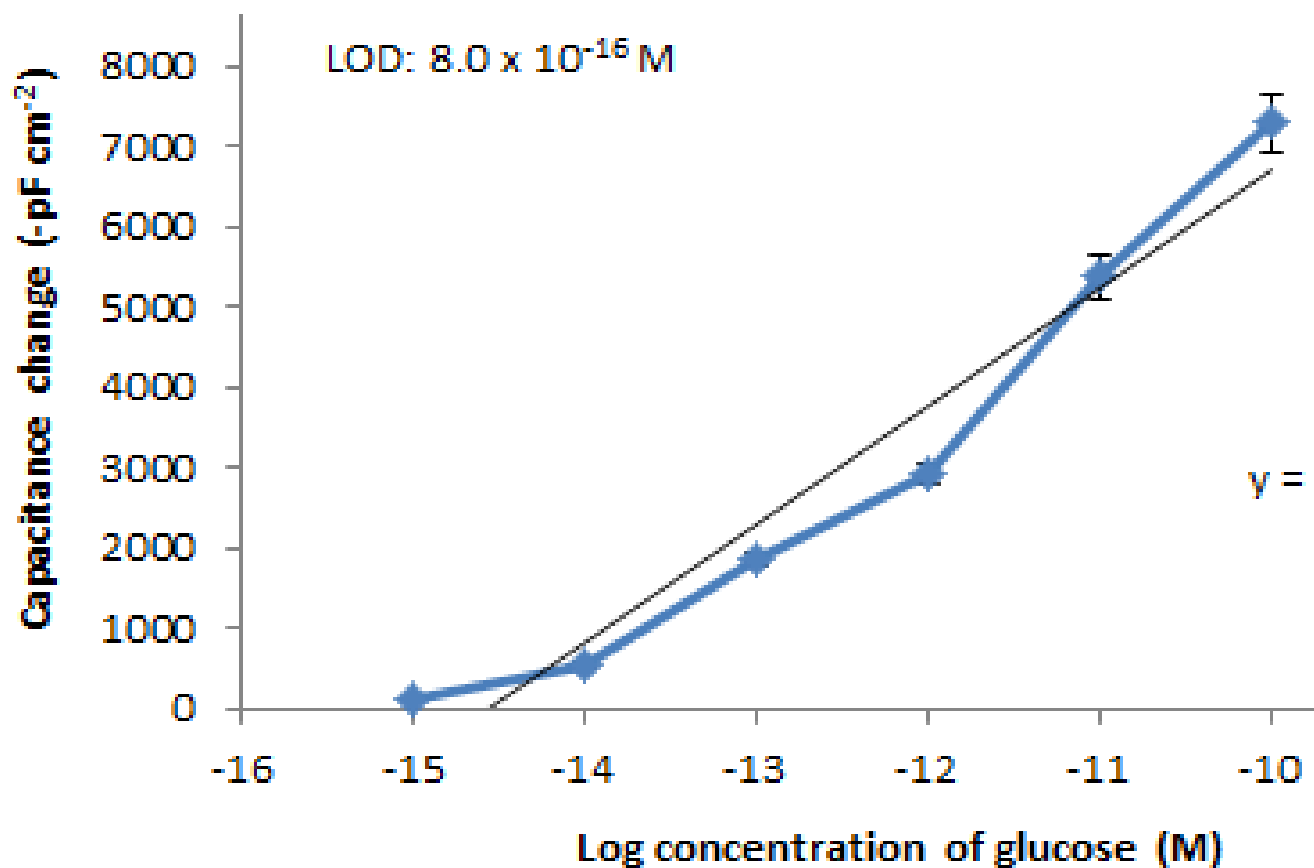


Figure 4. Calibration curve for glucose detection measured with immobilized APBA capacitive biosensor under optimum conditions (Flow rate $100 \mu\text{L min}^{-1}$; **sample volume 250 μL** ; running buffer 10 mM phosphate, pH 8.5; T 25 °C).

IgG detection using APBA electrode surface

Figure 5. IgG injection (10^{-12} M) into the immobilized APBA capacitive electrode under optimum conditions (Flow rate $100 \mu\text{L min}^{-1}$; **sample volume $250 \mu\text{L}$** ; running buffer 10 mM phosphate, pH 8.5; T 25°C).

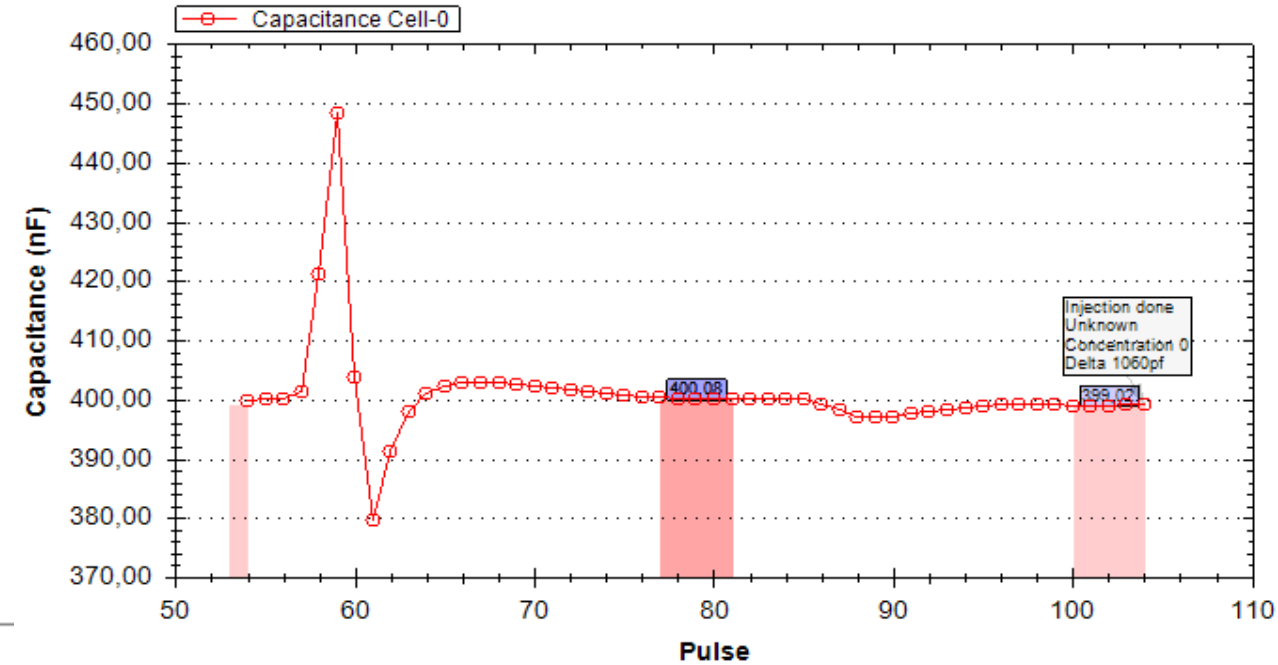


Figure 6. Calibration curve for IgG detection measured with immobilized APBA capacitive biosensor under optimum conditions (Flow rate $100 \mu\text{L min}^{-1}$; **sample volume $250 \mu\text{L}$** ; running buffer 10 mM phosphate, pH 8.5; T 25°C).

Evaluation of total capacity of electrode with immobilized antibodies

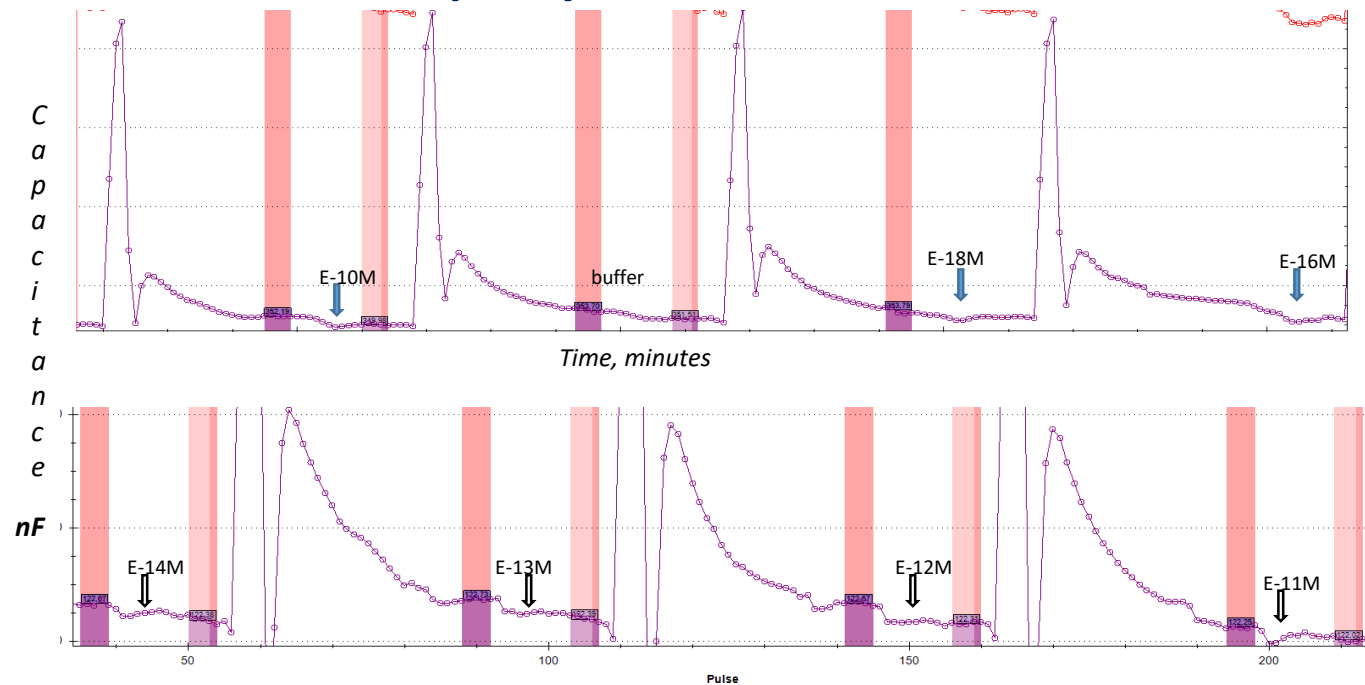


Figure 7. Response of the capacitance for electrode containing immobilized antibodies (antiTroponin T) on injections of regeneration buffer and analyte (Troponine T)

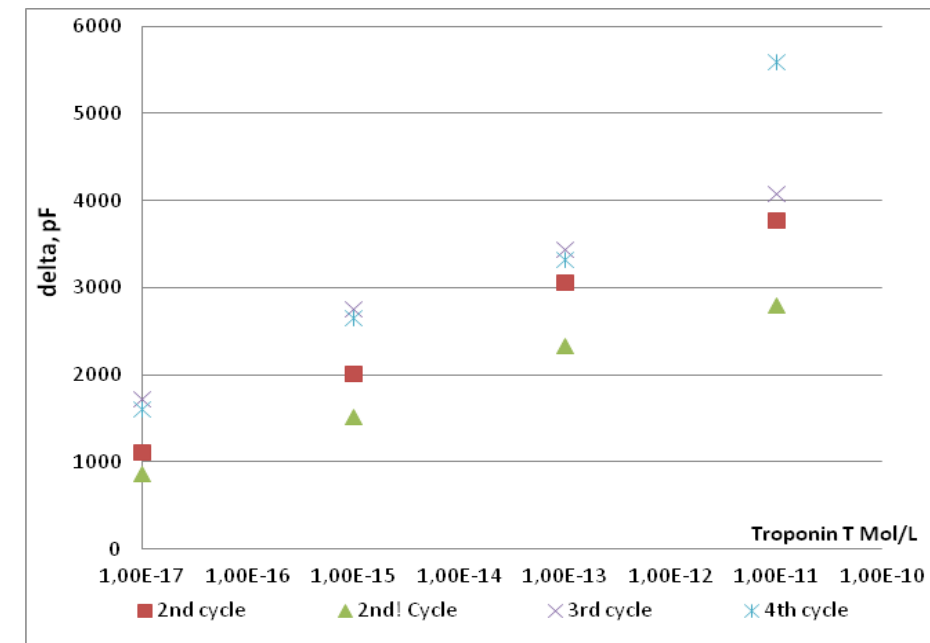
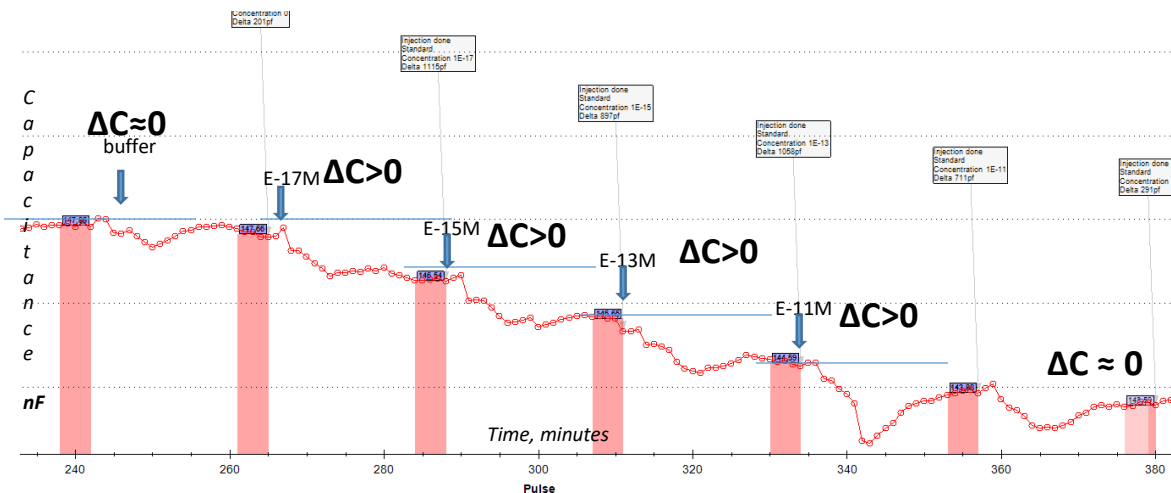


Figure 9. Calibration curve for Troponine T



$$\Delta C \text{ total} = \sum \Delta C = kN \text{ antibodies}$$

Figure 8. Response of capacitance for electrode with immobilized antibodies on injections of Troponine T, without regen. step

Reduction of nonspecific adsorption of proteins

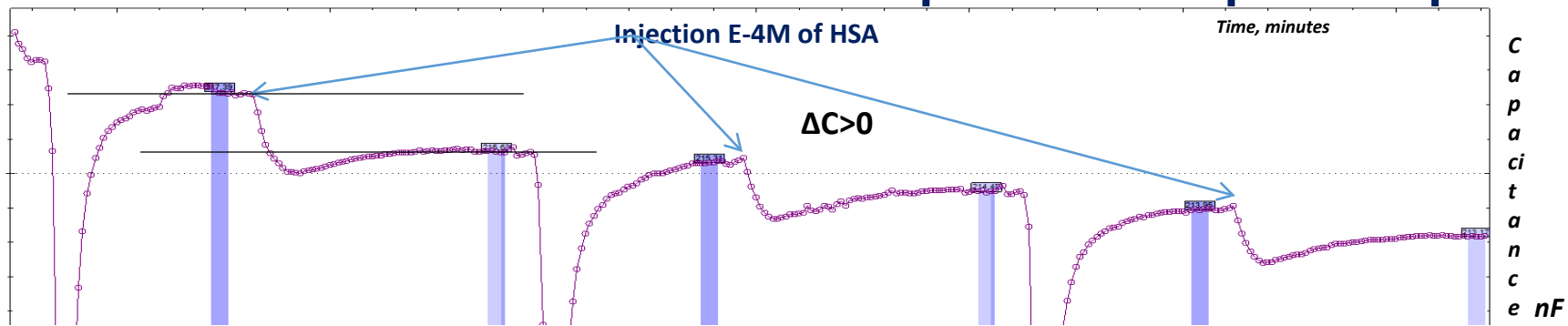


Figure 9. Response of the capacitance for electrode with immobilized antibodies and without PEG on injections of regeneration buffer (25mM glycine solution) and E-4 HSA.

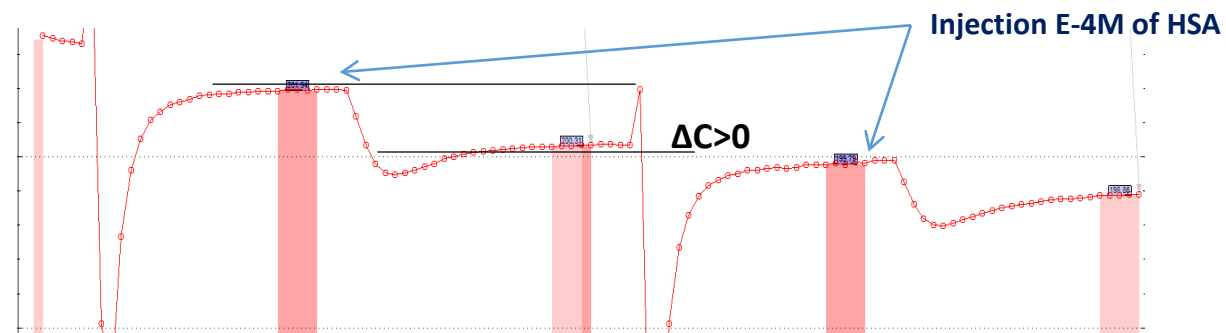


Figure 10. Response of the capacitance for electrode with immobilized antibodies and **PEG (350 Da)** on injection of E-4 HSA

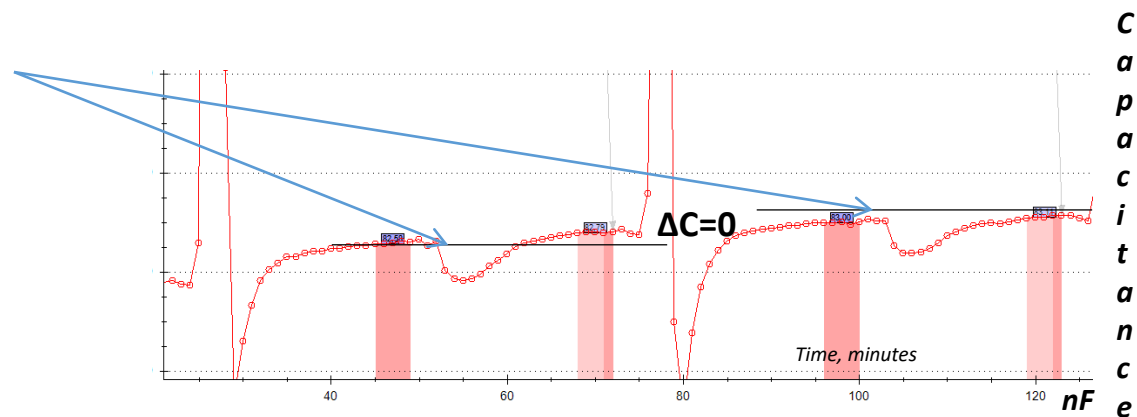


Figure 11. Response of the capacitance for electrode with immobilized antibodies and **PEG (500 Da)** on injection of E-4 HSA

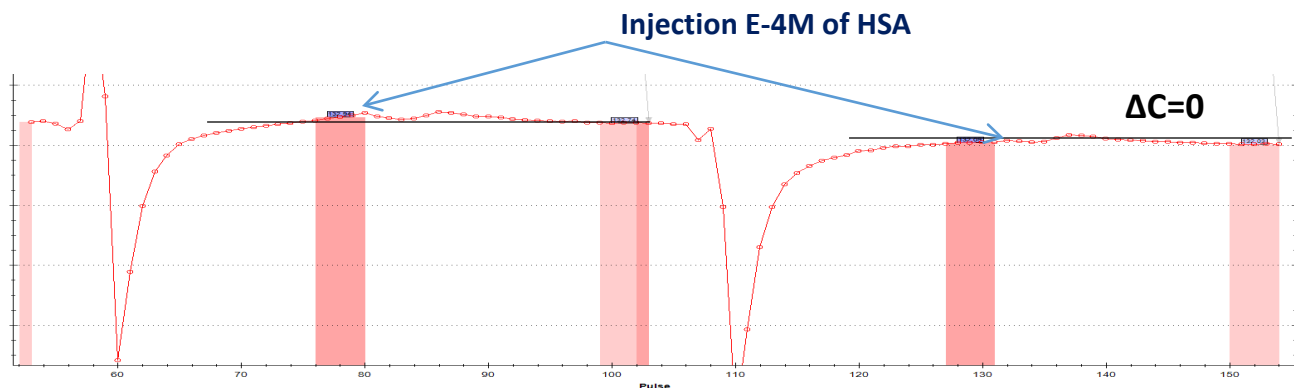


Figure 12. Response of the capacitance for electrode with immobilized antibodies and **PEG (950 Da)** on injections of E-4 HSA

Table 2. Immobilization strategy

Immobilization strategy	Contact time of mAb, sec	Self-assembled monolayer	Regeneration buffer 10mM glycine or NaOH, pH	Response, RU
2	900	DSP/AN	2.5	1052
3	900	DSP/AN	2.5	367.6
4	900	DSP/DMSO/streptavidin	2.5	277
12	450	DSP/AN	2.0	194
13	225	DSP/AN	11.0	345
14	125	DSP/AN	11.0	290
15	750	DSP/AN	11.0	596
16	750	DSP/AN DTT	11.0	978
17	750	DSP/AN CHI	11.0	311.6
18	750	DSP/AN CHI	2.5	311.6

RAPID DETECTION OF MYCOBACTERIUM TUBERCULOSIS USING SPR BIOSENSOR

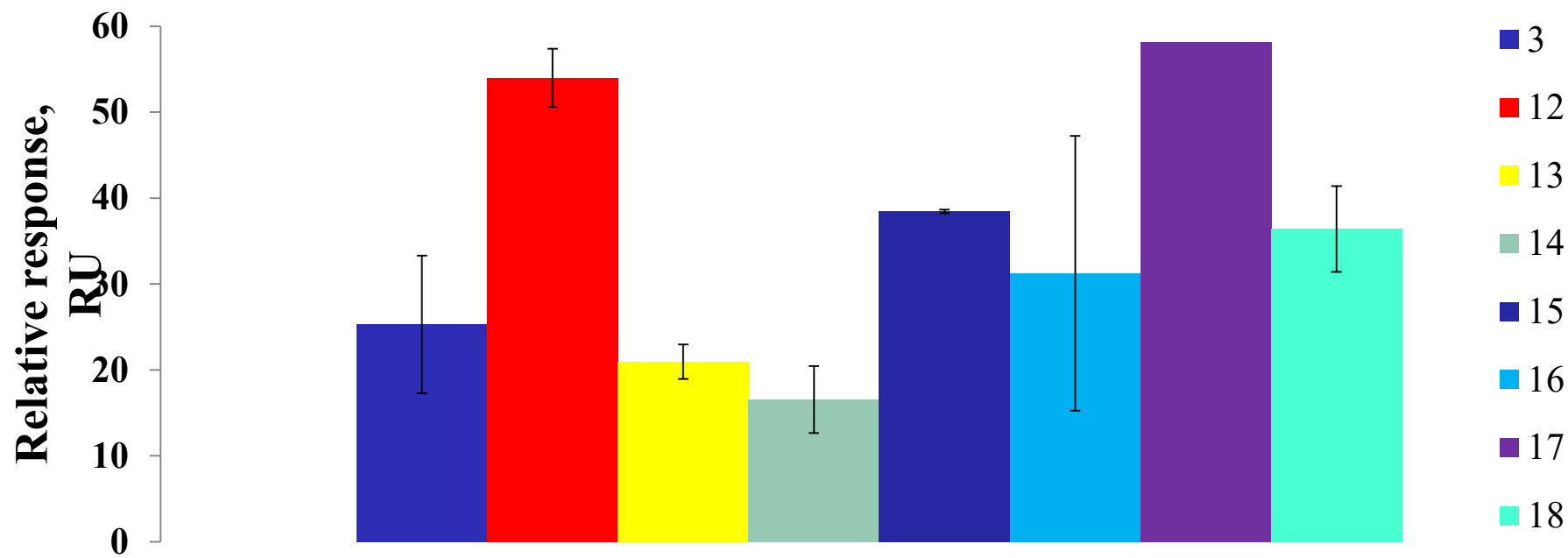
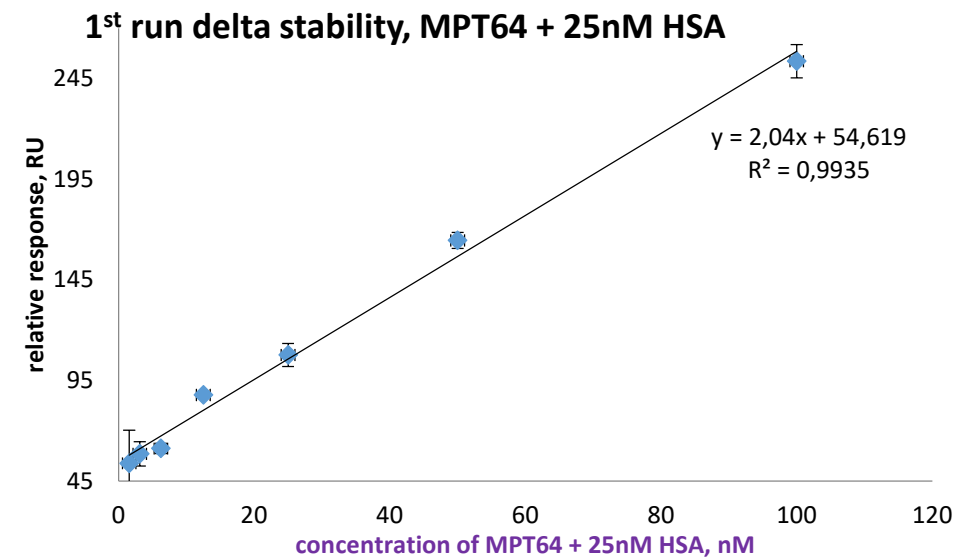


Figure 13. Delta stability response of electrodes with immobilized Ab MPT64 on injection of 25nM MPT64 at flow rate of 5ul/min.

Optimization of conditions of Ab MPT64 immobilization relatively response on the protein MPT64

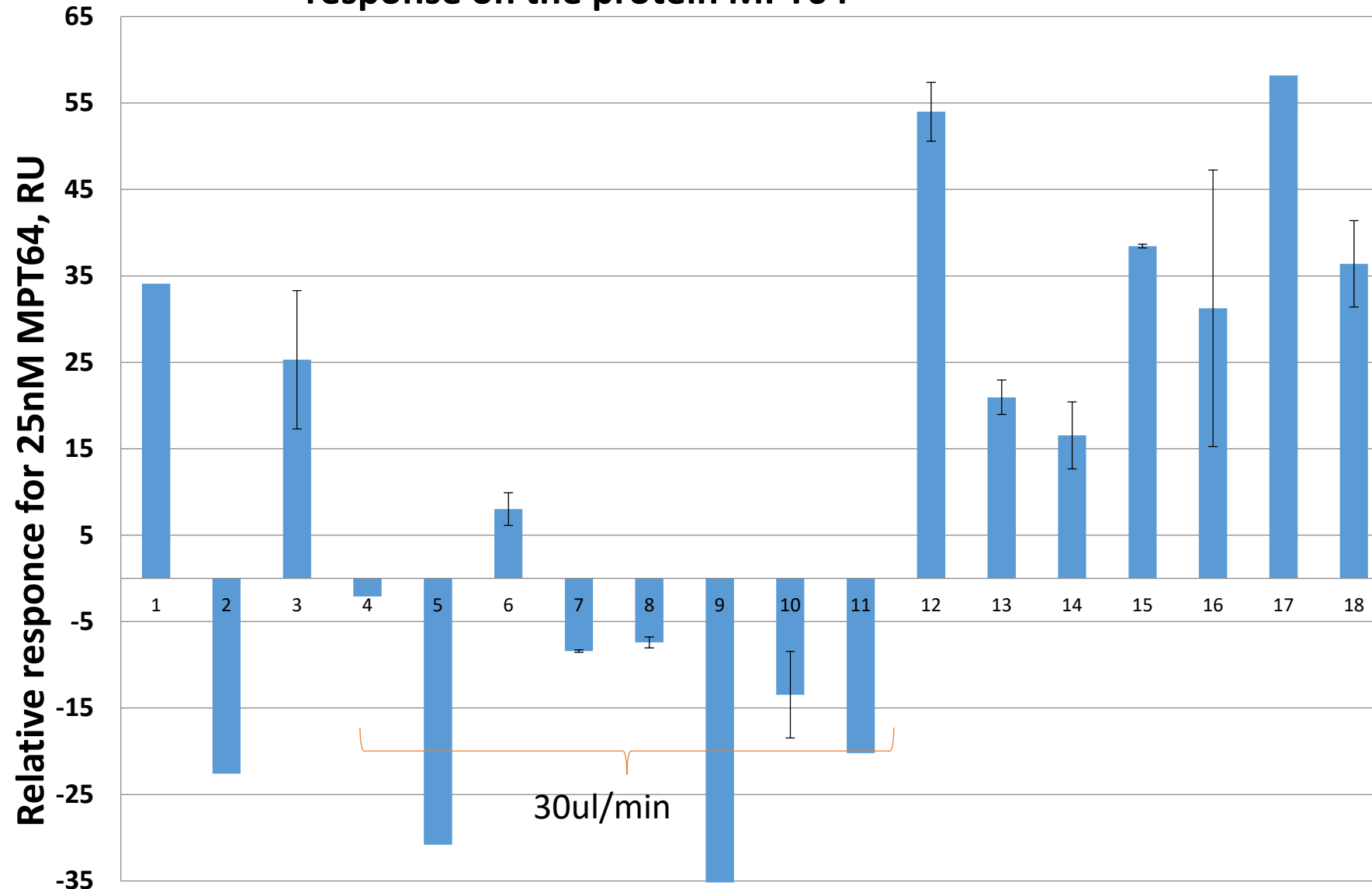


Figure 14. Delta stability response of electrodes activated using various methods with immobilized Ab MPT64 on injection of 25nM MPT64 at flow rate of 5ul/min and 30ul/min.