



Further sensitivity enhancement of HFF-QCM immunosensors for pesticides



C. March^{1,2}, J.V. García², R. Fernández², Y. Jiménez^{1,2}, A. Arnau^{1,2}, A. Montoya¹

¹Centro de Investigación e Innovación en Bioingeniería (Ci2B),
Universitat Politècnica de València.

²Advanced Wave Sensors, Valencia (Spain)



SUMMARY

Recently, High Fundamental Frequency Quartz Crystal Microbalance (HFF-QCM) immunosensors have successfully been developed [1]. Therefore, sensitivity of QCM biosensors is no longer a drawback. Taking advantage of this previous work, we have developed a renewed highly sensitive HFF piezoelectric immunosensor using carbaryl insecticide as a model analyte for pesticide detection. To this purpose, 100 MHz quartz crystal sensors were used as the transducer elements of the biosensor and a monoclonal antibody-based competitive immunoassay was integrated as the sensing specific bio-recognition event. The biosensing interface was improved by employing mixed self-assembled monolayers (mSAMs) of alkane thiols as intermediate layers for surface functionalization. This approach allowed the covalent attachment of the assay conjugate (20.0 µg mL⁻¹ of BSA-CNH conjugate) onto the gold electrode surface in a more orderly and stable way than with simple SAMs. A very low concentration (1.0 µg mL⁻¹) of LIB-CNH45 monoclonal antibody was used for the competitive immunoassays. All immunosensor assays were performed in the AWS A10 test platform from AWSensors. In terms of analytical performance, the new carbaryl HFF-QCM immunosensor showed higher sensitivity than the previously developed one [1], with analytical parameters very close to those of the most sensitive reported ELISA for carbaryl [2].

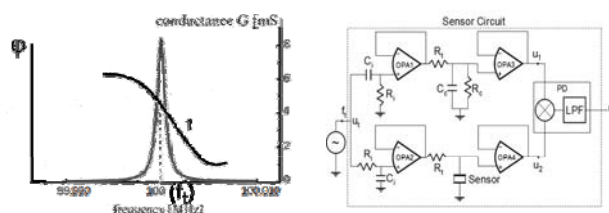
References:

[1] Carmen March, José V. García, Ángel Sánchez, Antonio Arnau, Yolanda Jiménez, Pablo García, Juan J. Manclús, Ángel Montoya, 2015. Biosens. Bioelectron. 65, 1-8.

[2] Abad, A., Montoya, A., 1997. J. Agric. Food. Chem. 45, 1495-1501.

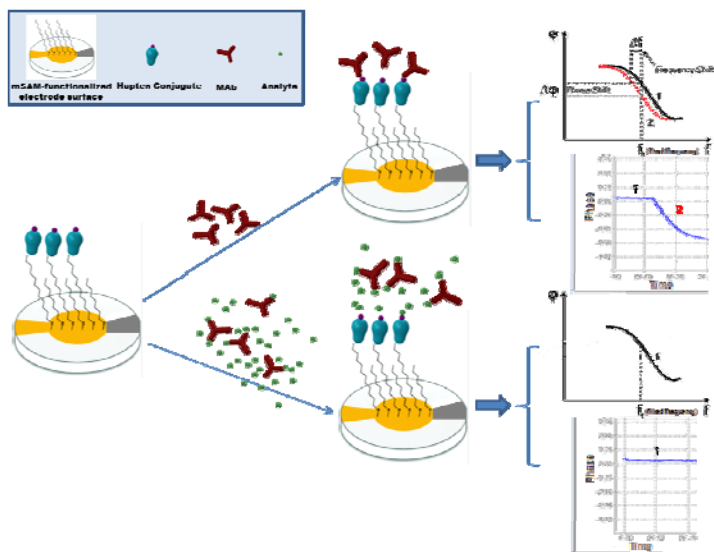
HFF-QCM IMMUNOSENSOR FUNDAMENTALS

New QCM sensor characterization technique based on the Phase/Mass sensitivity concept



$$\Delta\varphi = -\frac{\Delta m_c}{m_q + m_l}$$

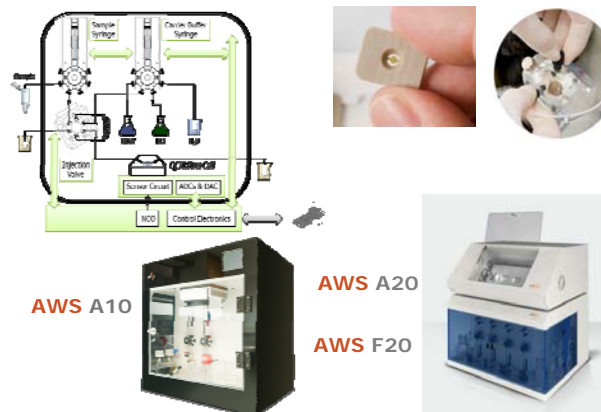
SENSING INTERFACE AND SIGNAL TRANSDUCTION STRATEGY



EXPERIMENTAL DEVICE

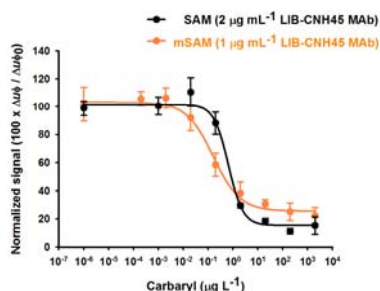
Scheme of the AWS A10 test platform from AWSensors

HFF-QCM sensor chip and flow cell assembly



RESULTS

HFF-QCM Standard Curve



Analytical Parameters

Analytical performance comparison between carbaryl HFF-QCM immunosensors (employing simple and mixed SAM) and ELISA.

Analytical parameters (µg L ⁻¹)	ELISA ¹	100 MHz HFF-QCM	
		SAM ²	mSAM
I ₅₀	0.06	0.66	0.16
LOD	0.01	0.14	0.01
LOQ	0.02	0.26	0.03
WR	0.02 – 0.18	0.26 – 1.72	0.03 – 0.90

¹ Abad and Montoya (1997)

² March et al., (2015)

REMARKS

- A renewed HFF-QCM immunosensor has been developed by using mixed SAMs as improved biosensing interface.
- It has been tested for sensitive detection of pesticides taken carbaryl as a model analyte.
- Its analytical performance greatly surpasses that of the previously reported HFF-QCM immunosensor for the same analyte. Moreover, the limits of detection (LOD) and quantification (LOQ), as well as the assay working range (WR), reach those of the most sensitive ELISA taken as the reference immunoassay.

ACKNOWLEDGEMENT

This work was supported by Spanish Ministry of Economy and Competitiveness project DETECTA-IPT-2012-0154-300000