A real winner for celiac patients

Universitat Autònoma de Barcelona

Tecan Award
Third place

The Tecan Award 2011 built on last year's success and saw our customers submit a plethora of innovative applications for our detection equipment. Competition was fierce, with many ingenious entries, and third place was eventually awarded to Maria Isabel Pividori from the Universitat Autònoma de Barcelona, for her work on the detection of gliadin, a constituent of the cereal protein gluten.

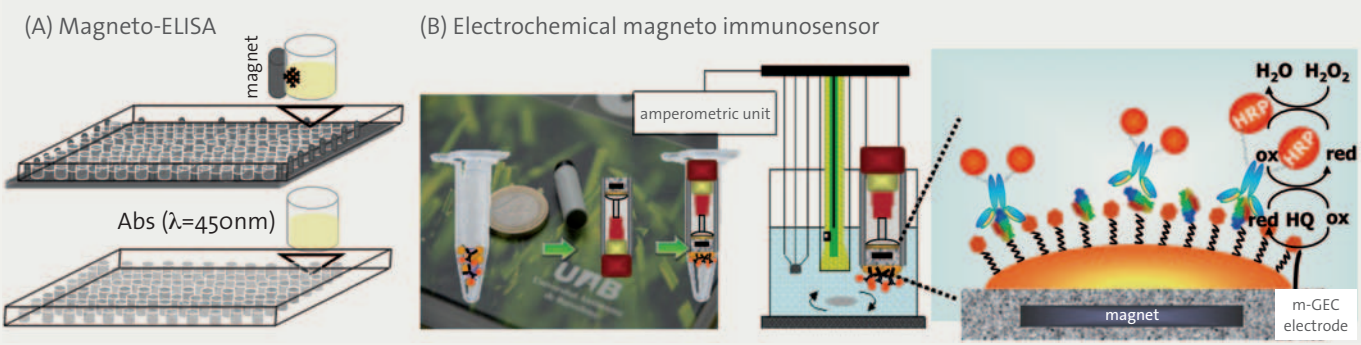
Celiac disease is the result of intolerance to gliadin, a constituent of the cereal protein gluten, requiring sufferers to strictly adhere to a lifetime gluten-free diet. A consequence of this is that the level of gliadin in gluten-free foods must be carefully controlled to ensure the food safety for celiac patients, making an easy and reliable method of gliadin analysis essential for food manufacturers producing gluten-free foodstuffs. To prevent celiac patients suffering adverse reactions to food, gluten is included in regulations relating to food labeling, with EC legislation stipulating that gluten-free products must not exceed a level of 20 mg/kg of food. Manufacturers must check for gluten contamination throughout the food production process and a key part of this is rapid testing of incoming raw materials.

Researchers in the Sensors and Biosensors Group at the Universitat Autònoma de Barcelona, Spain, have taken advantage of Tecan's Sunrise™ absorbance reader and Magellan™ V4.0 software to establish a novel magneto-ELISA for gliadin, based on optical detection. Maria Isabel explained: "There is an increasing demand for rapid, simple and low cost techniques for accurate food analysis in decentralized analytical situations. As a result, there is a great deal of interest in the development of biosensors for these applications, due to the high sensitivity and excellent specificity they offer. Immunosensors, in particular, are proving promising alternatives to the existing immunochemical tests that ensure food safety from the farm to the table."

"Recent advances have enabled magnetic beads to be used as a support for immunoassays. This significantly improves the performance of the immunological reaction, as the surface area is increased and the beads can be easily manipulated using a magnetic field. Assay kinetics are more rapid because the beads are in suspension, and the improved washing and separation steps help to minimize matrix effects in complex samples. In addition, the magnetic beads can easily be integrated into microfluidic devices and cartridges. We have taken advantage of this to develop an electrochemical magneto immunosensor for the sensitive detection of gliadin – and small gliadin fragments – in natural or pretreated foods such as skimmed milk and gluten-free beer. The immunological reaction is performed on magnetic beads

The Biosensors and Bioanalysis team. Left to right; Susana Liébana, Tamara Laube, Delfina Brandao, Soledad Carinelli, Daniela Santos, Rey Bundalian, Isabel Pividori

Experimental procedures



Schematic representation of experimental procedures for A) magneto-ELISA and B) electrochemical magneto immunosensor

as a solid support, enabling gliadin to be successfully immobilized and oriented onto the magnetic beads. The biorecognition strategy is based on a competitive assay – using commercially available antibodies labeled with horseradish peroxidase (HRP) – and the modified magnetic beads are captured onto the surface of a magneto electrode based on graphite-epoxy composite (m-GEC) for electrochemical detection. This low cost biosensor allows gliadin analysis to be performed easily throughout the food chain, enabling control of each stage in the production process."

Maria Isabel continued: "To assess the performance of this assay, we compared it with a novel magneto-ELISA, using optical detection performed on the Sunrise plate reader. As with the immunosensor, we immobilized gliadin on magnetic beads and used HRP as the label for the enzymatic reaction, optimizing both direct and indirect competitive assays. The ELISAs were performed in 96-well microplates, using a magnetic separation plate to isolate the supernatant before measuring the absorbance in the Sunrise reader. The recovery values using both methods were very good; the limit of detection for the magneto-ELISA was excellent and, as with the magneto immunosensor, we were able to detect levels of 20 µg/kg of food, which is 1,000 times lower than the EC-specified 20 mg/kg limit of detection for gluten-free food."

"I particularly like the versatility of the Sunrise reader," concluded Maria Isabel. "It enables magneto-immunoassays to be performed in a variety of different formats for multiple applications – such as evaluating protein coupling to magnetic beads and nanoparticles – as well as assessment of different transducer materials for biosensing purposes. It also allows us to quickly and easily optimize reagents and assay parameters, making it ideal for research applications."

Full details of this study can be found in: Laube T *et al*. Biosens Bioelectron, 2011, 27, 46-52.

To learn more about Tecan's Sunrise absorbance reader, visit **www.tecan.com/sunrise**

To find out more about Maria Isabel Pividori at the Sensors and Biosensors Group, visit **webs2002.uab.es/ipividori/**

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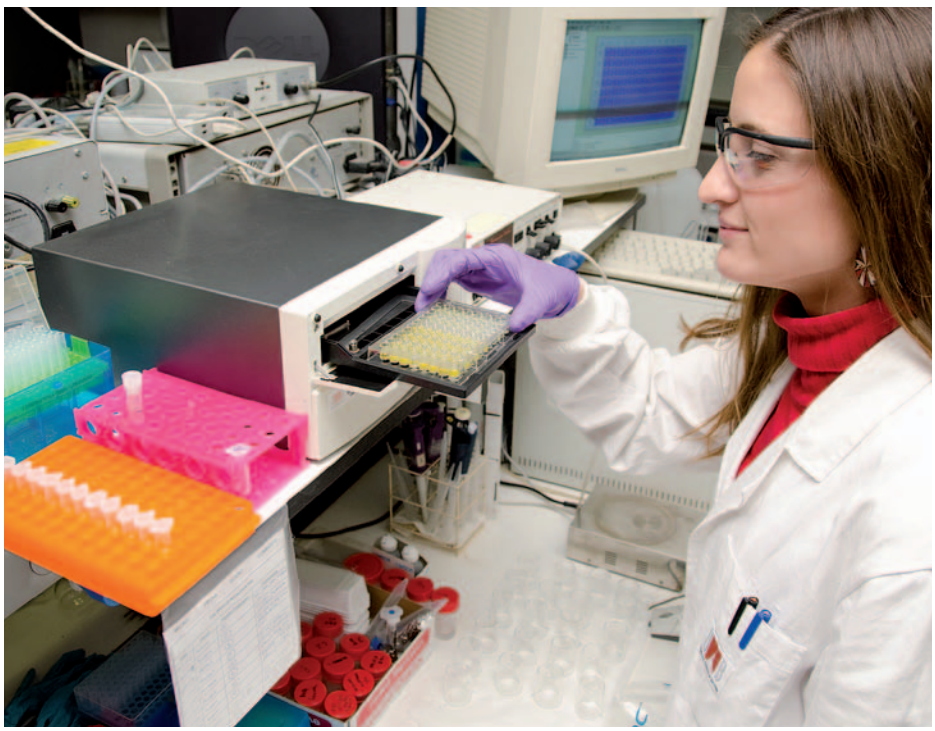

Tamara Laube inserts a plate into the Sunrise reader