Application Note

Bacterial Contaminants Detection in Drinking Water

3T analytik

Detection of bacteria in food, beverage and drinking water is crucial thus should be fast, sensitive and specific. QCM with antibody-mediated binding on sensors enables rapid real time Escherichia coli detection in drinking water, improving efficiency compared to conventional detection protocols

Summary

Detection of bacteria in food, beverages, and drinking water should be fast, sensitive, and specific. Within this context, microbial testing using the QCM device, qCell T, represents a useful approach for the rapid detection of contaminating bacteria.

Background

Mandatory detection of contaminating bacteria in food, beverages and drinking water is routinely performed according to recommended ISO procedures [1]. These techniques are relatively easy to perform but time consuming as cultivation-dependent approaches are often applied. Hence much effort has been undertaken to develop alternative rapid detection methods, which are as simple, specific and sensitive as the recommended techniques but able to deliver results within a single working day.

Strategy

To detect Escherichia coli (E. coli) in drinking water, the QCM sensor surface is equipped with an antibody that specifically binds to the bacteria. The QCM oscillates with a constant frequency when an aqueous solution, e.g. drinking water, is applied. If E. coli are present in the sample, the bacteria attach to the sensor surface, which in turn results in a decrease in resonance frequency. If bacterial counts are relatively low, the signal can be enhanced by applying antibody-coated nanoparticles, which bind on top of the bacteria, thereby further decreasing the resonance frequency. In this way, the presence of contaminating E. coli can be monitored in real time [2].
In this study the detection limit of the QCM device was determined. Figure 1 represents schematically the binding of bacteria and nanoparticles, respectively, to an antibody-coated QCM sensor.

Method

The surface of the QCM sensor is first treated with acetone, peroxymonosulfuric acid, and dried using nitrogen gas. The sensor is then coated with protein A and a polyclonal anti-LPS O- and K-antigen antibody. After a final washing step the sensor is ready to be used. At the start of each measurement a sterile sample is applied to calibrate the QCM device. Once the resonance frequency is stable, a bacteria-containing sample is applied and

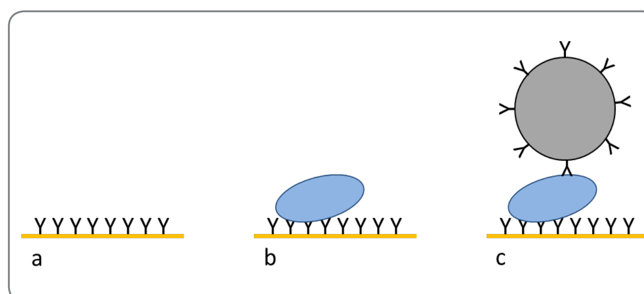

Figure 1. Schematic of an antibody-coated QCM sensor surface a) ready-to-use antibody coated sensor; b) bacteria specifically attached to the sensor surface; c) addition of antibody-coated nanoparticles to amplify the signal.

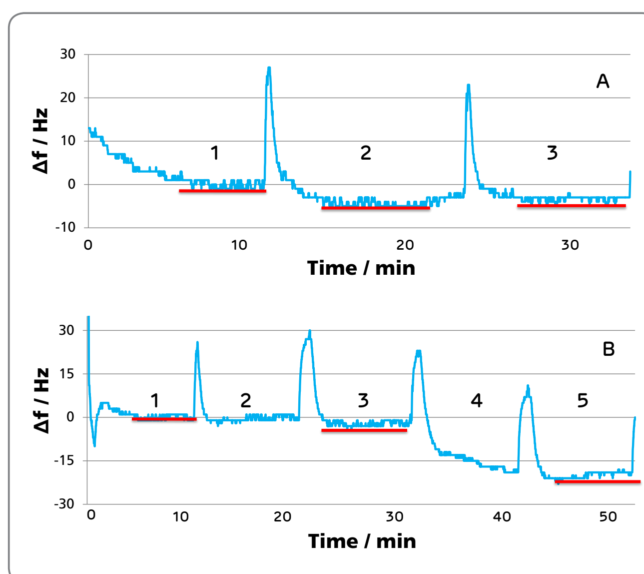

Figure 2. Detection of bacterial contaminants by resonance frequency changes. Panel A: Binding of bacteria to the sensor surface; Panel B: Signal amplification after the addition of nanoparticles to immobilized bacteria. 1: Calibration; 2: Application of $5x10^5$ bacteria/ml; 4: Signal amplification after the addition of $4x10^8$ nanoparticles/ml; 3 and 5: Confirmation.

changes in resonance frequency can be monitored. Figure 2A depicts typical resonance frequency changes when bacteria bind to the sensor. The frequency decreases approximately 5 Hz by applying $5x10^5$ bacteria/ml, which approaches the limit of detection. Therefore antibody-coated nanoparticles were applied to enhance the signal up to approx. 20 Hz, see Figure 2B. In addition, a control experiment was carried out with $4x10^8$ nanoparticles/ml non-antibody-coated nanoparticles, see figure 3.

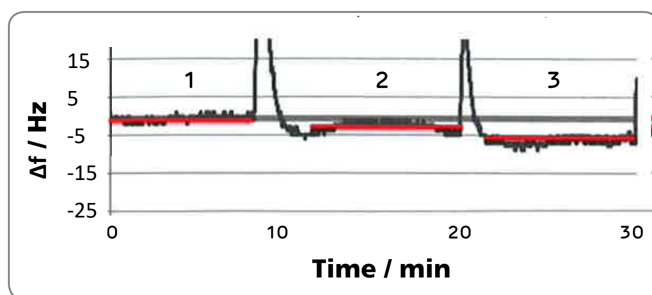

Figure 3. Control measurement with non-antibody-coated nanoparticles. 1: Calibration after immolizing $5x10^6$ bacteria/ml; 2: Application of $4x10^8$ bacteria/ml; 3: Confirmation.

Conclusion

QCM sensors from 3T analytik coated with antibody were used for $5x10^5$ bacteria/ml E. coli detection and nanoparticles were applied to amplify the corresponding signal. The qCell T instrument enables detection of E. coli in less than one hour in real time. This analytical method has the potential to be an alternative method to the conventional detection protocols in the future.

Reference

[1] http://www.iso.org/iso/catalogue_ics_browse?ICS1=07&ICS2=100&ICS3=30& ICS 07.100.30 "food microbiology"
[2] Jiang, X. et al. (2011) Evaluation of different micro/nanobeads used as amplifiers in QCM immunosensor for more sensitive detection of E. coli O157:H7. Biosensors and Bioelectronics 29: 23-28.

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3T analytik

3T analytik GmbH & Co.KG
take-off GewerbePark 4
D-78579 Neuhausen ob Eck

Tel.: +49 (0)7467-947-66-0
Fax: +49 (0)7467-947-66-29
E-Mail: info@3t-analytik.de
www.3t-analytik.com