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Nanowire technology for the detection of microorganisms in potable water

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Abstract

The lack of surveillance of the water supplies carry out a lot of epidemic issues supplies in developing countries ending in loss of human lives every year, being children the most affected age range. The aim of this study was surveying the microbial quality of water through the Electronic Nose (EN). The novel EN used in this study is equipped with an array of chemical gas sensor composed by 6 MOS (Metal oxide sensor), two of them fabricated using nanowire technology. This sensors array allows enhancing the threshold of the instrument in the detection of compounds in low concentration.

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Keywords: Nanowire; Water; Electronic Nose; Microbial Contamination; *Escherichia coli*.

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1. Introduction

One of the most common microbiological contaminants in water is Coliforms, being the *Escherichia coli* the representative microorganism of the group. In the case of developing countries the survey of the water supplies are not as accurate as it is required. This deficient controls trigger the appearance of epidemic issues where he most affected age range is children from 0 to 4 years. On the other and in the European Union has been establish by low the procedures to accomplish and accurate survey of the water supplies for the population. Although this control exists, every year in the EU are reported near 10000 of confirmed cases of *E. coli* toxins-producing as reported in the annual epidemiological report from the European Centre of Control Disease.

The methods used currently for the quality control of water require in most of cases a big amount of time and involves conventional techniques such as classical microbiology, chemical or molecular techniques. This time is not always available and it requires a big amount of money for the competent authority.

It is important to highlight that the microbial contaminant of water during their development develop complex metabolic pathways. A major part of these metabolites are volatile and are known as Volatile Organic Compounds (VOCs). The set of all the VOCs is characteristic of every specie of microorganisms. In this case it can be used like a fingerprint for the identification of the microorganisms and the concentration present in a solution. The aim of this work was the establish a new fouling based the use of a novel EN supported by classical techniques, like classical microbiology, for the detection of bacterial presence in water [1-3].

2. Materials and methods

2.1. Samples preparation

In a first step of the research a preliminary analysis where performed using microorganisms that are common contaminants in water, selected from the UMCC (UNIMORE Microbial Culture Collection). The selected microorganisms were *E. coli, Salmonella typhimurium* (data not sown) *and Listeria monocytogenes*. Separate cultures of all three microorganisms in BHI (Brain Heart Infusion) media [4]. Sterilized chromatographic vials (20 ml) containing 2 ml of BHI, where inoculated with 100 µl of the bacterial culture with a concentration of $9x10^8$ bacteria/ml. The analysis was carried out for 24 hours.

In the second step of the research a real sampling was done, isolating the group of coliforms from water of a water closet (WC) and a selected well. An aliquot of water from WC and a well was dispersed in Petri dishes with Violet Red Bile Agar (VRBA) (OXOID) [5]. VRBA is a selective medium used for the detection and enumeration of Coliform bacteria in water and other food dairy products. Selected colonies were transferred to BHI liquid media. Subsequently sterilized chromatographic vials (20 ml) containing 2 ml of BHI, where inoculated with 100 µl of the bacterial culture with a concentration of $9x10^8$ bacteria/ml. The analysis was carried out for 24 hours.

In both cases the control samples were performed just adding 100 µl of media instead of liquid culture to the 2 ml of BHI containing vials.

2.2. Electronic nose analysis

The EN used in this work was the EN EOS835 (SACMI IMOLA Scarl, Imola, Italy) is equipped with a thermally controlled sensor chamber of 20 ml internal volume where are placed 6 MOX gas sensors. Despite of this model of EN is on the market but the sensor array have been replaced in cooperation with the Sensor Lab CNR INO Brescia in Italy. Four of these sensors were prepared with the RGTO thin film technology [6], and the other two were constructed with nanowire technology [7]. Metal oxide nanowires sensors were prepared using the well-established evaporation-condensation technique, directly on the active transducers (Fig. 1). Electrical contacts and heaters were

deposited by RF magnetron sputtering. Devices were then soldered on TO package via gold wires and mounted in the EN.

In this way the adsorption surface is increased in a huge amount enhancing the response of the instrument and decreasing the threshold. It was also provided with the auto-sampler headspace system HT200 (HTA srl, Brescia, Italy), that allow to carried out a sustained analysis for 20 hours.

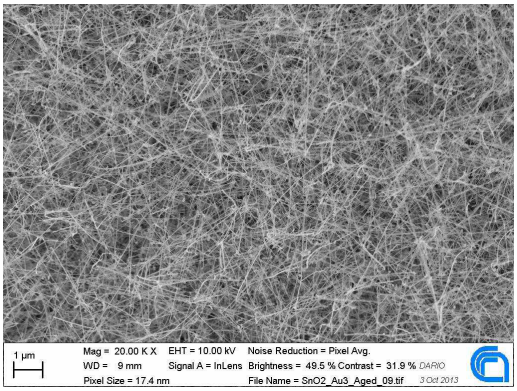

Fig. 1: SEM image of SnO_2 nanowires synthetized by evaporation-condensation technique.

The sensor baseline was performed by using synthetic chromatographic air. The data analysis was run by means of Principal Component Analysis (PCA), operated with the Nose Pattern Editor software (SACMI Imola Scarl, Imola, Italy).

3. Results and discussion

In the figure 2 it is possible to observe the PCA score plot from the analysis of *L. monocytogenes,* showing 3 well separated clusters. The first one is the cluster formed by butanol samples (internal standard of the instrument) (green triangles). The other two clusters make reference to the control samples (black stars) and the inoculated samples (blue dots).

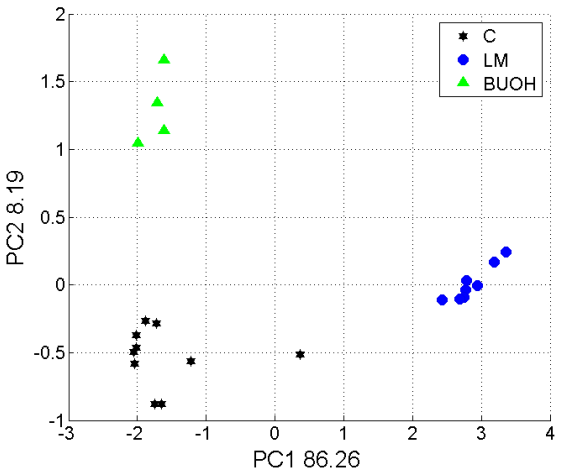

Fig. 2: PCA score plot showing the evolution of *L. monocytogenes* , during the first 24 h of analysis comparing with the control.

In the case of the inoculated samples it is possible to see that is well separated from the control cluster that determines a good recognition of the contaminated samples by the EN. Moreover the layout of this samples in the PCA space suggest that the instrument is able not only of identification the contaminated samples but also to individualize the different step of the microbiological curve of growth of *L. monocytogenes.*

Parallels results were obtained in the cases of the samples belonging to the microorganisms isolated from the water of well (Fig. 3). Control samples are well separated from the inoculated ones. As in the previous case it manifest a good recognition of the contamination by the instrument. It is possible to observe that the coliforms inoculated samples are aligned in the PCA score plot space following the steps of the growth curve. In this case the separation between the different inoculated samples is more evident than in the case of *L. monocytogenes*.

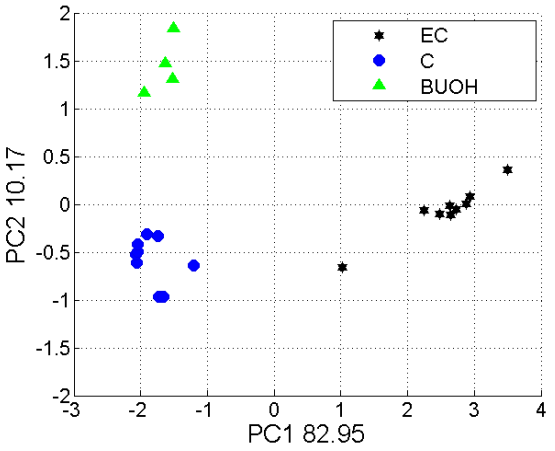

Fig. 3: PCA score plot showing the evolution of *E. coli*, during the first 24 h of analysis comparing with the control.

4. Conclusion

In conclusion it is possible to confirm that the EN equipped with the nanowire technology it's a fast tool to detect the contamination and to control the microbial development in water. Regarding this results the novel EN is an optimal candidate to be applied at line in industrial water survey process.

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