**Wearable sensor for real-time monitoring of oxidative stress**

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**1.Introduction**

The recent spread of SARS-cov-2 has shown that existing health systems are not capable of enabling the effective management of rapidly spreading infectious diseases. However, the limitations and critical points that emerged during the pandemic provide an opportunity for the formulation of more efficient models to reduce the transmissibility and mortality of infectious diseases. In particular, pathologies concerning airways, such as asthma, lung cancer and chronic pulmonary diseases are on the rise according to World Health Organization: in total, 3.9 million deaths each year worldwide are caused by respiratory diseases. A large proportion of respiratory diseases are chronic and require frequent check-ups to monitor their progression [1]. This work aims at developing an original methodology and an innovative, inexpensive and miniaturized device for monitoring patients suffering from respiratory diseases, through the non-invasive detection of hydrogen peroxide in respiratory exhalations. In particular, the attention was focused on the fabrication of an electrochemical sensor that can be inserted into normal face masks in order to detect the concentration of hydrogen peroxide in exhaled air. H2O2 is one of the Reactive Oxygen Species (ROS) used to evaluate the level of oxidative stress in respiratory system: it is the most used biomarker because it is more stable than the other ROS and it diffuses through the cell membrane so it can be monitored in the extracellular space. High levels of this metabolic product are related to the onset of oxidative stress. This condition is a risk factor for several diseases, and therefore its early detection can help in risk assessment, disease monitoring and prevention.Therefore, the use of an electrochemical sensor allowing real time monitoring of hydrogen peroxide in breath exhalate can provide additional information and it is cost-effective, rapid and non-invasive compared to traditional diagnosis methods that are not suitable for a wearable continuous monitoring [2].

**2. Methods**

The sensor was realized from the silver layer present on the compact discs (CDs) and the three electrodes configuration was given through the use of a laser cutter [3]. An AgCl paste was applied to act as reference electrode and a graphite layer was applied to the counter electrode, while the Ag working electrode was not modified. In addition, electrospun polylactic-acid membrane (PLA) or an electrodeposited chitosan layer was used as absorber material. The application of this layer facilitates the wettability of the sensor in presence of a humid atmosphere such as that given by exhaled breath (relative humidity close to 100%). To study the performance of sensor, electrochemical tests (Linear Scan Voltammetry) were carried out in liquid solution at different concentrations of hydrogen peroxide between 10 µM and 5mM. These solutions were prepared by dilution of H2O2 in 0.1 M PBS (Phosphate Buffered Saline) at pH=7.4. Then, these tests were repeated using aerosol of hydrogen peroxide solution in order to simulate humid atmosphere of exhaled breath [4].

**3. Results and discussion**

LSV tests were performed by placing the sensor in the liquid solutions at different concentrations of hydrogen peroxide and applying a variable potential between -0.2V and -0.9V vs Ag/AgCl. The obtained current density was proportional to the amount of hydrogen peroxide present in the solutions. The detection of hydrogen peroxide in the aerosol phase was carried out by exposing the sensor to the nebuliser for a time sufficient for homogeneously wets of the surface layer constituted by PLA membrane or electrodeposited chitosan. The electrochemical detection was performed with the same parameters used for liquid solutions. The experimental results showed that both layers are able to absorb the humidity of the aerosol and that the sensor is also able to quantify the hydrogen peroxide concentration in the aerosol phase.

**4. Conclusions**

In this study we reported the development of a wearable sensor for the detection of hydrogen peroxide in exhaled air since high levels of this metabolic product are related to the onset of oxidative stress. In order to facilitate the wettability of the sensor in presence of a humid atmosphere such as that given by exhaled breath a PLA membrane or a chitosan layer was used. The sensor developed was tested using hydrogen peroxide solution in a concentration ranging from 10 µM to 5mM in liquid phase and in aerosol phase in order to simulate humid atmosphere of exhaled breath. LSV tests showed that current density was proportional to the amount of hydrogen peroxide present in the solutions and that the sensor is also able to quantify the hydrogen peroxide concentration in a humid atmosphere.

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