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Impediometric Technique as a Rapid Estimation to Detect Pathogenic Bacteria in Quality Control

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It is of interest for drug- and food-producing industries to acquire information about the microbiological quality of their products as early as possible. Conventional microbiological methods for the detection and identification of pathogens include microbiological culturing and isolation of the pathogens, followed by approbation with biochemical and serological tests. These methods are time-consuming and labor-intensive, not suitable for a rapid response for high-risk pathogens. As a result, several instruments have been developed for use much earlier, including nucleic acid-based tests, serological methods, and some automated instrumental diagnostic assays (1).

The impedance measurement is used as a rapid method to detect microbial growth. It is based on a measurement of the changes in electrical conductance of the culture media that are produced by microbial metabolism. Impedance is the resistance to flow of an alternating current through a conducting material. The examined microorganisms are cultivated in impedance glass tubes (cells) in fluid growth medium, with highly uncharged molecules that are fitted with measuring electrodes. The culture medium is metabolized by microbial activity, and the impedance changes and decreases because of the production of small charged molecules by the microorganism's metabolites (2). The impedance measurement is a complex entity composed of a combination of a conductive element and a capacitive element. Therefore, in monitoring microbial growth, conductance measurements can indicate changes taking place in the bulk solution or medium (3). The specific growth medium used in the impediometric technique supports the target bacteria, and must be formulated to provide optimal impedance signals to provide specificity to the impedance microbiological method (2).

Impedance measurements are based on the bacterial growth and metabolic processes that produce electrically measurable changes in the growth medium. These changes are due to the metabolism of high-molecular-weight nutrients into smaller charged ionic molecules, which increase the electricity conductivity of the specific medium. Therefore, the impedance technique distinguishes viable and dead pathogens rapidly, within 24 hours (4). Variation in electrical conductivity is related to the change in bacterial number, and thus bacterial growth can be measured. Accordingly, impedance measurements are considered for quality control in the food, drug, and hygiene-product industries, especially for the identification and enumeration of indicator microorganisms (2, 5).

In glass tubes, the electrical change is recorded using a pair of electrodes immersed in the growth medium. The impedance measurement can be done by two methods: direct or indirect. In the direct method, a pair of metal electrodes is submerged in the medium that is inoculated with the testing sample. The electrical change caused by the release of ionic metabolites from live cells in the medium is monitored over time (6). In the indirect method, the electrodes are immersed in a separate solution (usually a potassium hydroxide solution) instead of the inoculated growth medium. The gases (mainly CO_2) released from the metabolism of bacteria are absorbed by the potassium hydroxide solution, which leads to decreased conductance of the alkaline solution. The indirect method is applicable to all microorganisms, including yeasts and molds (2, 6).

In the impedance technique, by means of an electrode system in the glass tubes, impedance values can be recorded over time for a pre-selected incubation period (7). The concentration of ions generated by the bacteria reach a magnitude at which a measurable increase in conductivity can be detected. The bacterial level associated with this change in conductance is called the bacterial threshold level, and the impedance detection time (IDT) is defined as the point in hours. Usually, the IDT is inversely propor-

tional to the log number of bacteria in the sample. Therefore, bacterial counts can be estimated by measurement of the IDT. Finally, at the termination of the incubation periods (24 and 48 hours), the printouts of the impedance patterns and the IDTs expressed in hours for each test cell are produced (2, 7). Usually, IDT is not visible until the number of bacteria reaches approximately 10^6-10^7 CFU/mL. Finally, when it reaches a concentration of 10^8 CFU/mL or greater, the impedance value shows a plateau. In this time, all of the resources in the medium have been metabolized into end-products, and the shape of the impedance growth curve is mainly associated with the usual bacterial growth phases. Many studies indicate the detection time is related to the initial cell concentration (2). Impedance techniques in microbiology have been used for the detection of many bacteria, as well as yeasts and molds in various samples. This technique also can investigate the growth behavior of microorganisms. Studies have shown that the impedance growth curves under different conditions are related to bacterial species' characteristics (2, 6, 8, 9).

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