Proceedings of the 2nd International Conference on Biological Engineering and Medical Science (ICBioMed 2022), Part I
DOI: 10.54254/2753-8818/3/20220363

Application Of Different Spectral Methods For Target Analysis

Yue Fu[1,5,†], Jinghan Zhang[2,†], Shi Zhen[3,†] and Yuhaohua Zheng[4,†]

[1]College Alpin Beau Soleil, Vaud, Villars sur ollon 1884, Switzerland;
[2]Xi'an Gaoxin No.1 High School, Xi'an 710075, China;
[3]School of Science, Xi'an Jiaotong-Liverpool University, Suzhou 215123, China;
[4]Ningbo Foreign Language School, Ningbo, 315000, China

[5]yue_fu@beausoleil.ch
[†]These authors contribute equally.

Abstract. Spectral analysis methods are widely used for the analysis of different targets such as viruses, bacteria, heavy metals and their harmful substances due to their simplicity and efficiency. Among these spectral analysis methods, the study of luminescence and light absorption of different substances has important theoretical and practical significance. Atoms are different and emit different bright-line spectra, and the atoms of each element have a certain bright-line spectrum. Therefore, a diverse of different spectroscopic techniques can be used to achieve various target detection. In this research, four common spectral analysis methods are introduced, including Raman spectroscopic analysis, electroluminescence analysis, fluorescence analysis and colorimetric analysis. And this research analyzes the detection effect of these four spectral analysis methods on different targets, such as virus and pathogens. With the features of scientific type spectroscopic instruments or systems with high sensitivity, high resolution, high reliability and multidimensional information in spectroscopic techniques, the developed spectroscopic techniques to detect viruses effectively. And the demand for its application will be more urgent and will surely become a direction of great interest for quite a long time to come.

Keywords: Nanomaterials, Spectral Analysis, Detection.

1. Introduction

Spectral analysis methods are a kind of technology that based on optical signal to detect selective expression system of chemical and biological components. They use the spectrum of the sample (such as absorption, reflection, fluorescence and chemiluminescence) to analyze the interesting information. The introduction of nanomaterials in spectroscopic analysis methods can make the practical application of analytical methods even better, because nanomaterials show quantum size effect and macroscopic quantum tunnel effect. And relying on their excellent optical properties, electrical properties and good biological compatibility, nanomaterials-based spectral analysis methods have been widely used in biochemical analysis, in gene regulation, drug screening and the application of small molecule and protein detection. For example, spectral analysis methods constructed with metal nanomaterials can be used to analyze a diverse of different target and show high sensitivity and selectivity. As a result, spectral

© 2026 The Authors. This is an open access article distributed under the terms of the Creative Commons Attribution License 4.0 (https://creativecommons.org/licenses/by/4.0/).

DOI: 10.54254/2753-8818/3/20220363

analysis methods have been applied in different fields such as environmental analysis, catalysis and energy [1].

Based on the research on spectroscopic techniques, four common spectroscopic detection techniques have been developed [2], namely fluorescence, Raman spectroscopy, colorimetric sensing method and electrochemiluminescence (ECL). For fluorescence analysis, the fluorescence protein can be modified to suit the properties of the application and in the detection of fluorescent proteins. Most of them are genetically modified because of sulfide-reactive functional groups, which make them more sensitive. This high sensitivity allows it to detect single molecules with ease. For ECL analysis, the ECL is one of the most commonly used receptors by using different nanomaterials, such as AuNPs. Changing the nanocomposite in this way can enhance the stability and efficiency of ECL emission, which is more conducive to the detection of the analyte. For colorimetric sensing method, by using noble metal nanoparticles as sensing elements, the detected substance can be identified and confirmed by the color change, and the performance of the developed colorimetric analysis method is further improved, which can facilitate different target detection. In general, the four common spectral analysis methods described above have been widely used. As a result, this study mainly introduces the application of these four spectral analysis methods in different fields.

2. Raman spectroscopy

There is only one nucleic acid in a virus: DNA or RNA, which is required for it to reproduce in a replicative manner in order to be differentiated from other life forms. Viruses are currently a big threat to humans, with untold numbers of people being infected by different viruses every day, especially with the current ravages of the new crown and monkeypox viruses. Viruses can cause damage to different systems in the human body, for example, the nervous system, digestive system, and respiratory system. Sub-gingival infections and lesions are the main causes of invasive periodontitis. Usually, the number of several microorganisms around the teeth of periodontitis patients is more than the normal oral cavity. Therefore, it is necessary to find a way to effectively detect specific virus to anticipate the invasive periodontitis.

The presence of sub-gingival human cytome galovirus (HCMV) and epstein-barr virus (EBV) was associated with invasive periodontitis [3-4]. With previous precedents regarding the detection of other viruses, the HCMV virus that causes invasive periodontitis can also be detected by Raman spectroscopy. The pp65 is one of the HCMV tegument proteins, which is a low matrix phosphorylation protein. It is not only a major antigen of HCMV viremia, but also a major target protein of HCMV-specific cytotoxic T cells. The EBV belongs to the herpesviridae family, herpesvirus type I V, which is a human lymphocytic herpesvirus. EBV infection is very common in the human population. It has been found that more than 90% of the adult serum EBV antibody positive. Because (EBV) is very susceptible, it can be transmitted through saliva, droplets, blood transfusion, etc. The EBV can infect and transform B cells through the adsorption of related viral proteins on the surface of B cells. Once infected, EBV is difficult to remove and will be carried for life. After infection with EBV, the body will produce corresponding antibodies against different antigens, such as anti-VCA-igg. By detecting these EBV-specific antibodies, it is clear that they are primary EBV infection or previous EBV infection or reactivation of previous EBV infection. Since lgM is the earliest antibody after antigen stimulation in the body, we can detect the presence of lgM in saliva by using SERS. Through machine learning of saliva of different normal people in the early stage, it can be further analyzed. If it is found that the results obtained from the spectrum are not consistent with the normal values learned before, early intervention can be achieved in the early stage of periodontitis.

Similar to the putative detection of novel coronaviruses, a chemical reaction may occur after HCMV enters an agent, and it is here that Raman spectroscopy can take place. Herpesviruses and bacteria may play a synergistic role in the pathogenesis of invasive periodontitis. To better accomplish the goal of detecting HCMV and EBV with sensitivity and reliability, Raman Spectroscopy could be utilized. Raman spectroscopy, the displacement of the wavelength of light by using the interaction force resulting from the inelastic part of the interaction between light and molecules is called the Raman shift. It is this

DOI: 10.54254/2753-8818/3/20220363

shift that gives each molecule its own "identity". With the help of machine learning, different viruses can be recognized by the computer, being used as a comparing group when Raman spectroscopy is used to detect various viruses. Raman spectroscopy makes molecules highly specific to some chemicals. However, since the Raman signal is very weak, 2-D materials such as transition metal dihalides are needed to generate additional signal enhancement, a method called surface-enhance Raman spectroscopy (SERS), which is highly sensitive and specific, as demonstrated in a previous paper on breast cancer.

Shi et al. have recently designed a SERS-based breath analysis module that can screen for new coronaviruses in less than 5 minutes (Fig. 1), which is described in the article as a superior protocol to nasopharyngeal swab and polymerase chain reaction (PCR) detection. The breath analysis module contains a chip carrying three sets of SERS probe molecules, which can be used to attach the nanomaterials. When the subject exhales into the device for 10 seconds, the new coronavirus biomarkers in the exhaled breath react chemically with the sensors. The breath analyzer is then loaded into a portable Raman spectrometer, and the reacted compounds are then characterized according to the change in SERS signal [5]. The SERS-based sensors and machine learning technology can quickly and accurately detect novel coronavirus, and results can be obtained in only 25 minutes, with an accuracy of up to 92% as compared to the accuracy of PCR testing. This sensor does not require sample preparation or complex manipulation skills, and is particularly suitable for large-scale population detection, offering a strong advantage over existing test methods. However, its manufacturing cost is expensive, and few laboratories will use SERS to detect microorganisms, so the popularity of SERS is not as wide as that of qPCR, and a lot of knowledge stays in few scientific research directions and uses SERS technology. We also expect that the more new-generation SERS technology can make up for the shortage of this technology and become a cheap and effective detection technology.

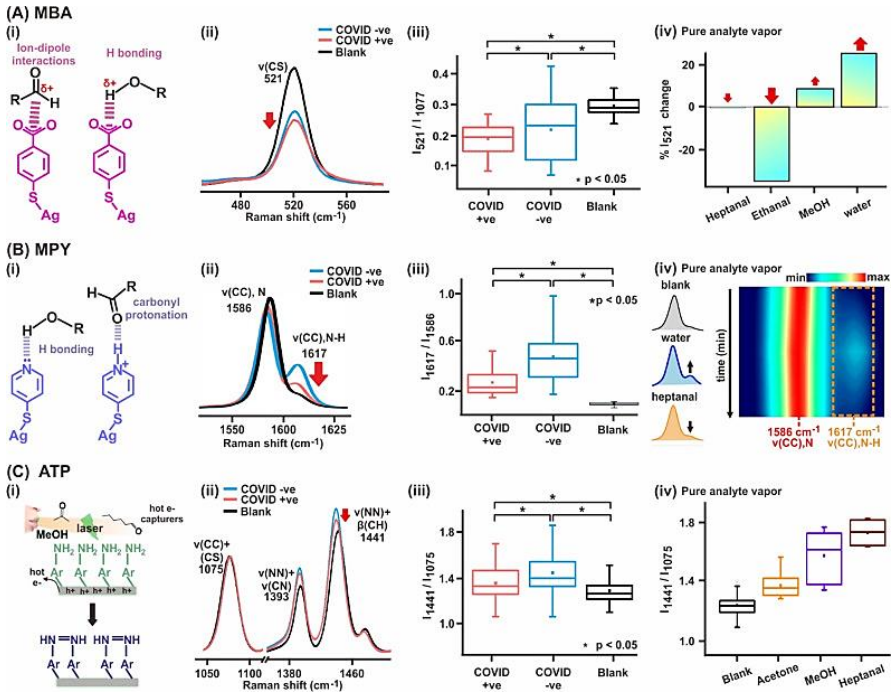


Figure 1. The analysis results for the breath samples by using the SERS analysis method (5).

3. Fluorescence analysis

Oral diseases are one of the most well-known diseases in dentistry and the world health organization suggesting it is one of the most important public health issues now days. The two main oral afflictions known as dental cavity and periodontal diseases. Oral diseases can be caused by many micopathogens

DOI: 10.54254/2753-8818/3/20220363

and regular oral examination can help detecting these diseases earlier as some of them might restrict the daily activities in our life even lead to oral cancer [6].

Lactobacillus was known to be one of the first pathogen related to dental caries and periodontal diseases. It is known as a bacterium causing lactic acid after carbohydrate fermentation leading to an acidic environment. Saliva is a mixture differs by individual containing 99 percent of water and 1 percent of electrolytes and proteins. Lactobacillus covers only 0.1 percent of saliva. But there are research showing correlation between the amount of lactic acid and dental plaque. However, the formation of cavity and periodontal diseases caused by lactobacillus is not clearly identified. Some research suggest that lactobacillus is an acid tolerance bacterium an survive in the acidic environment below 4.5. As lactobacillus produces lactic acid after carbohydrate fermentation. They could alter the neutral oral environment to below 4.5, which enhances the development of oral diseases.

Periodontal is caused by a variety of pathogens attaching to the surface of teeth. The host has a complex relationship of cellular and molecular with the host tissues. Microorganisms closely associated with periodontal diseases are porphyonas gingivalis, bacteriode intemedius, actinobacillus actionmycetemcomitans and eikenella corrodens. This disease is one of the main reasons causing tooth loss. Researchers are trying to find the best method for early detection and diagnoses in order the reduce the damage of periodontal disease. The patients who has prior evidence of periodontal disease radiographic assessment is used to provide information for clinical examination for bone loss. One of the most important approaches is to find a sensor that is quick, selective, and sensitive that can be used within the Clinique for periodontal diseases.

Fluorescence is one of the is one of the most important methods used in medicine science. Fluorescence plays one of the main roles of diagnosing periodontal disease. Fluorescence can be used detecting neopterin within the gingival crevicur fluid. This method only requires small preparation and small number of analytes is required. The measuring principles are that when an electron absorbs light, it is taken up to an excited electronic state. The excited molecule will rapidly drop down and lose energy to the surroundings as it drops down to the lowest vibrational level of the excited electronic state. After this process, the excited state can undergo spontaneous emission and emit the rest of the energy as light. Therefore, giving a result of the detection wavelength longer than emission wavelength. Plaque sample of the patients taken and placed in a microtiter plate. The bound and unbound florescent are separated and the total bound florescent is detected using fluorimeter. The advantage of fluorescence is that it is sensitive and can detect single molecules. The instructions are simple and can be easily applied in clinics. The general idea of using bacteria concentration florescence immunoassay (BCFIA) is that the modified bacteria containing the sample of the patient. The procedure is performed within a fluricon assay plate [7].

Fluorescence protein is commonly used in clinics for biological imaging. Its property allows it to maintain the structure and localization of the target object. Fluorescence protein can be altered and engineered to a property suitable for specific detection and applications. Most Fluorescence protein are genetically modified using the sulfide-reactive azide functional groups. This property allows them being more sensitive with fluorescence. It was also taken into consideration whether examination requires part mouth or full mouth. Clinical examination tends to start with gingival and periodontal tissues by looking at the inflammation. Another example of using florescence to help in detection of curative cancer. DeLong et al. developed a new fluorescence laparoscopy model, which allows real-time fluorescent labeling of target tumors in mice [8]. With epithelial ovarian cancers the receptor folate receptor-α (FR-α) is often overexpressed. In addition, the folate-FITC can be used to selectively label ovarian cancer cells. The imaging system coordinates with this protein helping to visualize the image is the real-time multispectral intraoperative fluorescence imaging system. This system helps identifying tumor that are within the length of one millimeter. In order to improve the cancer detection and treatment procedures, a variety of techniques have been discovered selectively labelling cancerous cell with florescence molecules. Carriers developed a more advanced sophistication that are able to detect fluorescently label cancer cells as well as having the ability to destroy the remaining microscopic cancer.

DOI: 10.54254/2753-8818/3/20220363

4. Electroluminescence analysis

The electrochemiluminescence (ECL) can be used in different fields, such as pathogen analysis, heavy metal detection and disease analysis and tracking. Generally speaking, it can emit chemiluminescence by electron transfer reaction. With the advancement of technology development, the ECL technology has the characteristics of high anti-interference stability and specificity by change the size of the surface area to make it bigger and electronic conductivity of the electrode surface. There are usually two methods for ECL detection: ion annihilation pathway and co-reactant pathway, as shown in Fig. 2. First of all, the first method is about the generation of electrochemistry, which requires the participation of cationic and anionic, which will reach the surface of the electrode through the emitter. The two ions with opposite charges will react to generate ground state and excited state. Among them, the excited state will generate a light with a special wavelength, so that the ground state can be achieved while the ECL is generated. Compared with the first method, the co-reactant pathway is obviously more complex in process, and this method is closely related to the emitter and co-reactant. This ECL is generated because the anode or cathode potential is added to the solution containing the luminophore and co-reactant, which can change the polarity of the potential, the electrode surface will have oxidized or reduced luminophore and co- reactant, this is the process of generating radical ions and co-reactant species [9].

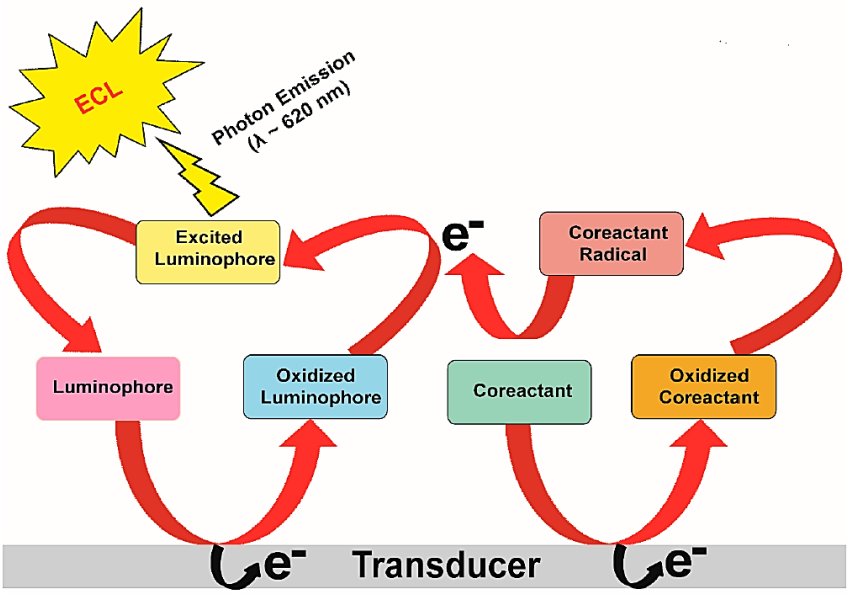


Figure 2. Schematic diagram of the mechanism of ECL (9).

In recent years, it can be found that ECL technology is growing slowly. Through understanding, it can be found that it is one of the current research trends, which can be mainly divided into four categories, such as ECL aptamer sensors, ECL immunosensors, ECL enzyme sensors and ECL DNA sensors. The immunosensors and the enzyme sensors are highly used. In the future, the development of this technology will make it more convenient and widely used in people's lives, especially in the field of biomedicine. The four categories of techniques also differ in their application to analysis. For example, ECL immunosensor, which combines immunoassay technology with electrochemical sensing technology, this sensor has good specificity, sensitivity and response speed, and can be used to detect trace substances. The other is called ECL enzyme sensor, which has special organic macromolecules, so this enzyme is characterized by high selectivity and catalytic activity, and can also be monitored in real time. And most of them can be used in diseases, food hygiene and environmental testing. In order to improve its performance, graphene, a substance with good electron transfer ability, has been tried to be used in new research, which may lead to new composite materials with improved performance.

DOI: 10.54254/2753-8818/3/20220363

ECL biosensor has the characteristics of high specific recognition function, high sensitivity, selectivity, fast response speed, convenient operation, etc. These advantages allow it to have a good development in recent years, and will achieve more in the future high accuracy and portability, in order to facilitate the operation and use of the effect [10]. There are some special ECL biosensor appear when the use of material and advances in the technology. The conductivity of electrons has been improved, allowing its biosensors to improve in performance. Gold nanoparticles is also one of the more widely used sensors based on metal and magnetic nanocomposites. The current research focuses on using its specificity to target specific biosensor applications. Due to its unique properties and functions, it can be well used in nanocomposite materials, because the increase of electrode surface area makes the new ECL sensor developed by it have high stability and sensitivity. Enhanced electrical conductivity for improved sensitivity. In addition, silver nanoparticles also have certain functions, such as promoting the transfer of electrons, which can make them have higher strength, thereby increasing the ability of some properties. As shown on the chart, it can be used to improve the ECL signal. In general, they all improve part of the performance through nanomaterials, making biosensors have more prominent advantages.

5. Colorimetric analysis

Colorimetric analysis is a method that uses the change color of the solution after adding colorimetric reagents or nanomaterials to observe and compare the color depth of the solution with eyes, or uses photoelectric colorimeter to measure the concentration of the substance to be measured in the solution. Ions of different valence states of an element have a specific color, which its depth is related to the concentration of the ion. The proportional relationship between the color of the ion and the concentration in the solution can be used to compare and analyze the concentration of the ion in the solution. Colorimetric analysis is simple, rapid and sensitive, and is widely used in the determination of trace components. With the help of colorimetric biosensing, which does not apply complex actions and high cost, people can detect the color changes in point-of-care diagnosis and on-site analysis without the help of machines.

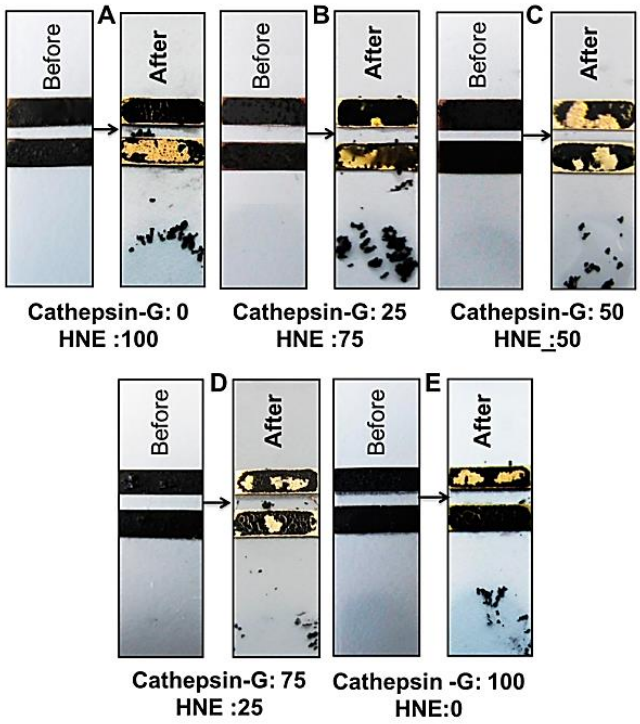


Figure 3. The developed colorimetric method for the analysis of periodontitis (11).

DOI: 10.54254/2753-8818/3/20220363

Periodontitis is a kind of inflammation caused by dental plaque, odontolith, traumatic occlusion, and so on. Periodontitis has been confirmed by the medical community as the third major killer of human health after cancer, cardiovascular and cerebrovascular diseases, and the "top killer" of oral health. Some of the early symptoms of periodontitis, but also become the main characteristics of oral "sub-health" deterioration. As a result, the detection of periodontitis has received increasing attention. For example, Wignarajah et al. developed a colorimetric sensing method for the analysis of typical biomarker for periodontitis [11]. In this work, they used specific proteases as probes to analyze proteolytic activity. By introducing magnetic nanoparticles, one end of these probes can be covalently bound to the magnetic nanoparticles and the other end to the surface of the sensor. As shown in Fig. 3, the sensor turns black when the structure is intact. When proteolyzed, the sensor surface leaks gold. Based on this color change, the detection limit of this colorimetric analysis method can reach 100 fg/mL and 1 pg/mL for Cathepsin-G and Human Neutrophil Elastase, respectively.

6. Conclusion

In summary, this research mainly analyzes four common spectral analysis methods for the analysis of different targets, including virous, periodontitis and cancer. Overall methods introduced in this essay are Raman spectroscopy, ECL and fluorescence, and as well as colorimetry measuring the color of the solution before and adding the reagent. The main diseases that have been introduced is one of the most common oral diseases worldwide- periodontal disease. Investigating different detection method can help us recognize and diagnose viruses quicker and easier to be applied in clinical uses.

References

[1] Harris, C. D., Kemp, G. E., Schneider, M. B., Widmann, K., Rubery, M. S., & May, M. J. (2021) Spectral sensor error analysis for measuring x-ray radiation drive using the DANTE diagnostic toward inertial confinement fusion experiments. The Review of scientific instruments, 92(3), 033505.

[2] Zhou, H., Liu, J., Xu, J. J., Zhang, S. S., & Chen, H. Y. (2018) Optical nano-biosensing interface via nucleic acid amplification strategy: construction and application. Chemical Society reviews, 47(6), 1996-2019.

[3] Sugano, N., Ikeda, K., Oshikawa, M., Idesawa, M., Tanaka, H., Sato, S., & Ito, K. (2004) Relationship between Porphyromonas gingivalis, Epstein-Barr virus infection and reactivation in periodontitis. Journal of oral science, 46(4), 203-206.

[4] DING Fang, FENG Xiang-hui, MENG Huan-xin, ZHAO Yi-bing, ZHANG Li, LU Rui-fang, CHEN Zhi-bin. (2008) Relationship between herpesviruses and periodontal pathogenic bacteria in subgingival plaque. Journal of Peking University (Health Sciences). 03, 318-322.

[5] Leong, S. X., Leong, Y. X., Tan, E. X., Sim, H., et al. (2022). Noninvasive and Point-of-Care Surface-Enhanced Raman Scattering (SERS)-Based Breathalyzer for Mass Screening of Coronavirus Disease 2019 (COVID-19) under 5 min. ACS nano, 16(2), 2629-2639.

[6] Petersen P. E. (2003) The World Oral Health Report 2003: continuous improvement of oral health in the 21st century--the approach of the WHO Global Oral Health Programme. Community dentistry and oral epidemiology, 31 Suppl 1, 3-23.

[7] Vernerová, A., Krčmová, L. K., Heneberk, O., Radochová, V., & Švec, F. (2022) Liquid chromatography method with tandem mass spectrometry and fluorescence detection for determination of inflammatory biomarkers in gingival crevicular fluid as a tool for diagnosis of periodontal disease. Journal of pharmaceutical and biomedical analysis, 212, 114644.

[8] DeLong, J. C., Hoffman, R. M., & Bouvet, M. (2016) Current status and future perspectives of fluorescence-guided surgery for cancer. Expert review of anticancer therapy, 16(1), 71-81.

[9] Rizwan, M., Mohd-Naim, N. F., & Ahmed, M. U. (2018). Trends and Advances in Electrochemiluminescence Nanobiosensors. Sensors, 18(1), 166.

[10] Ying Chen, Shiwei Zhou, Lingling Li, Jun-jie Zhu (2017) Nanomaterials-based sensitive electrochemiluminescence biosensing. Nano Today, 12, 98-115.

DOI: 10.54254/2753-8818/3/20220363

[11] Wignarajah, S., Suaifan, G. A., Bizzarro, S., Bikker, F. J., Kaman, W. E., & Zourob, M. (2015) Colorimetric Assay for the Detection of Typical Biomarkers for Periodontitis Using a Magnetic Nanoparticle Biosensor. Analytical chemistry, 87(24), 12161-12168.