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Review

The rapid detection of pesticide residues in fruits and vegetables using electrochemical methods

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Abstract. To meet global demand, pesticides play a very important role in the control of pests that damage agricultural products. However, overuse of pesticides threatens both ecosystems and human health. In recent years, gas chromatography (GC), liquid chromatography (LC), gas chromatography-mass spectrometry (GC-MS), liquid chromatography-mass spectrometry (LC-MS), capillary chromatography (CCH), and capillary electrophoresis (CE) have been used to determination of pesticides. Although these methods have high specificity, selectivity and sensitivity for detection of pesticides, they also have some shortcomings, for example, expensive instrument, sample prepared process is complex and time-consuming. Therefore, electrochemical sensors and biosensor have become good analytical methods for the detection of pesticides due to their many advantages such as simple manufacturing process of the detection system, high sensitivity and selectivity. In this paper describes the latest results used for in situ detection of pesticide residues in fruits and vegetables obtained in the fields of cyclic voltammetry, square wave voltammetry, differential pulse voltammetry and electrochemical impedance spectroscopy. In addition, the development of electrochemical methods and real-time smartphone monitoring techniques and their integration with detection platforms are discussed. For readers with a scientific and technological focus, this article will provide additional valuable information for understanding the creation of portable electrochemical devices, rapid detection of pesticides, the role of electrochemical sensitive methods and contributing to their further development.

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Introduction

Pesticides are synthetic or natural compounds or additives used to control, destroy, regulate, prevent diseases, weeds, insects and pests that affect plant growth. These compounds can often be categorized by their chemical structure, mode of action, hazard level, route of application, and time of application (Figure 1). Crop yields have increased significantly over the past decades due to the introduction of chemical pesticides. The global demand for pesticides has increased dramatically and steadily since the mid-1940s, mainly due to commercial farming [1, 2]. However, excessive and uncontrolled use of pesticides has led to contamination of food and environment, agriculture and water resources. Pesticide waste is becoming a serious public health problem due to penetration into food through fruits, vegetables, processed foods, water, air and soil, and acute and chronic effects on human health. Chemical pesticides can have cytotoxic, mutagenic and carcinogenic effects on human health [3, 4].

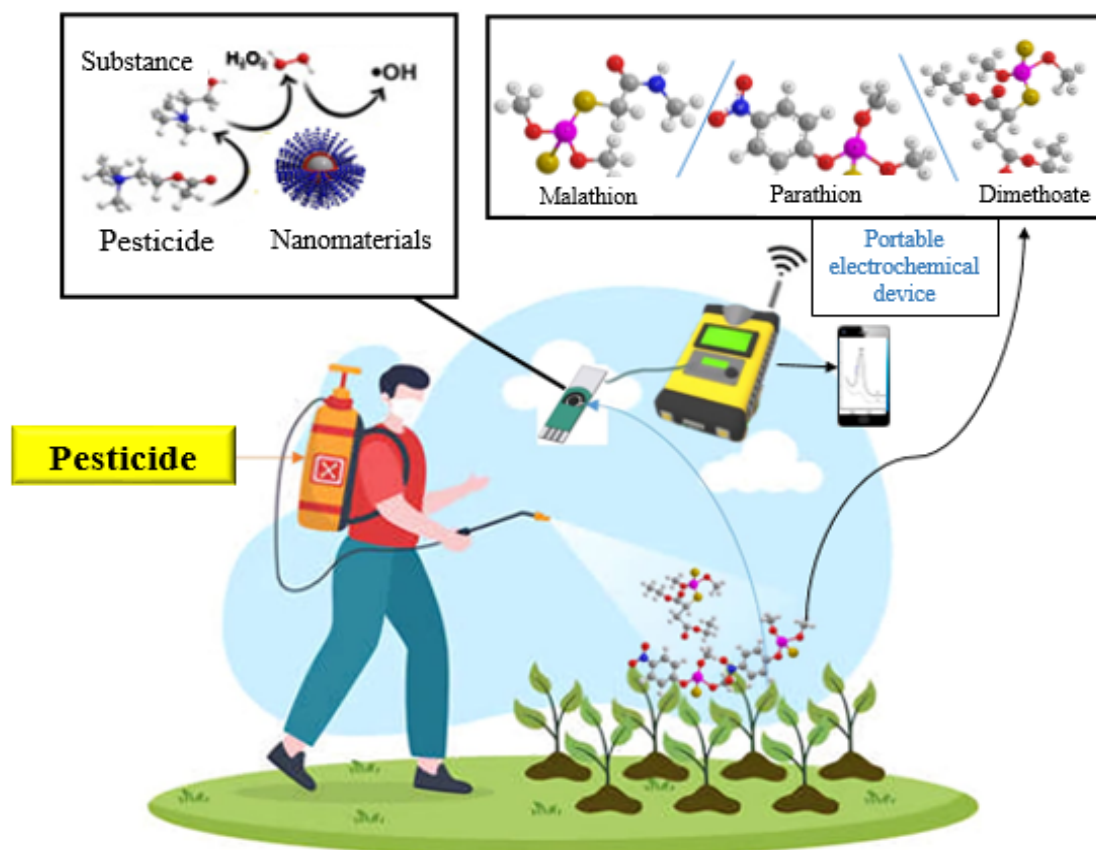


Figure 1. Schematic plan of pesticide determination

Many papers discuss laboratory-scale pesticide detection methods for the effective detection of adulterant residues and hazardous chemicals in food products. For example, Kim *et al.* developed aptamer-based fluorescent methods for the detection of fipronil using carboxyfluorescein-labeled aptamers [5]. The analysis showed that linear labeling of fipronil in 25-300 ppb range with limit of detection (LOD) of 53.8 ppb (10^{-7}) and a recovery rate of 94.7-114.4%. Yang *et al.* and many others wrote about methods of fluorescent sensor based on cadmium telluride quantum dots (CdTe QDs) for detection of residues of pesticide methidathion ($C_6H_{11}N_2O_4PS$) in Chinese cabbage with the limit of detection 0.027 ng/mL [6].

Capoferri A.D. *et al.* [7] illustrated similarity-based sensory approaches for screening pesticide residues in food products. In particular, they focused on optical (surface plasmon resonance, fluorescence, surface-enhanced Raman scattering and colorimetric) and electrochemical (amperometric, voltmetric, impedance and potentiometric) aptasensors, immunosensors and molecular polymer sensors. Tsagkaris A.S., Pulkrabova J., Hajslova J. [8] considered the achievements and new trends in optical methods for monitoring pesticide residues in food products, Chen K. *et al.* summarized the advances in the field of organophosphorus pesticides (OPs) and colorimetric and fluorescent sensors for the detection of their substances in the nervous system. Patel H., Rawtani D., Agrawal Y.K. [9] published articles on new trends in the development of chitosan-based sensor platforms for organophosphorus pesticides using acetylcholinesterase (AChE), particularly detection methods for various pesticides (colorimetric, potentiometric and amperometric), transducers combined with chitosan and modifications of AChE and can be effectively used for the detection of catalytic and biocatalytic organophosphate hydrolases using in situ detection devices. For hydrolysis reactions, square wave voltammetry has additional selective parameters compared to amperometric sensors for organophosphorus compounds at a given potential. And Weston M., Geng S., Chandrawati R. [10] investigated the opportunities and challenges of replacing nutrition sensors with more sophisticated models based on the latest advances in modern electronic and optical nutrition sensors targeting pesticides, humidity, gases, pathogens, temperature and pH. The development of electrochemical methods and the integration of smartphone technologies with electrochemical sensor platforms for real-time monitoring were also discussed.

Pérez-Fernandez B., Costa-Garcia A., Muniz A.E. [11] screen-printed discussed electrodes (SPEs) as an analytical (bio)sensor for electrochemical detection of pesticide residues for food traceability. Wang W. *et al.* [12] reviewed the advances in the development of electrochemical biosensors for pesticide identification with emphasis on the use of nanomaterials and molecular biology. H. [1] demonstrated the use of biomolecules, 3D printing and smartphone-based electrochemical biosensors for the detection of organophosphorus compounds. All these studies were conducted in laboratory conditions and are based on the development of electrochemical sensors.

Literature review. Electrochemical analysis methods are relatively low-cost and simple equipment and are versatile and powerful approaches that provide accuracy, high sensitivity and selectivity. Electrochemical sensing is based on the measurement of electrical quantities such as potential, current, or charge that result from the interaction between an electrode and the substance being analyzed. Typically, an electrochemical cell consists of a counter electrode (CE), a working electrode (WE) and a reference electrode (RE). In voltammetric methods, a time-dependent variable voltage (for example, Ag/AgCl) is used on the reference electrode (RE) and the current response between WE and CE is measured, where a redox reaction occurs.

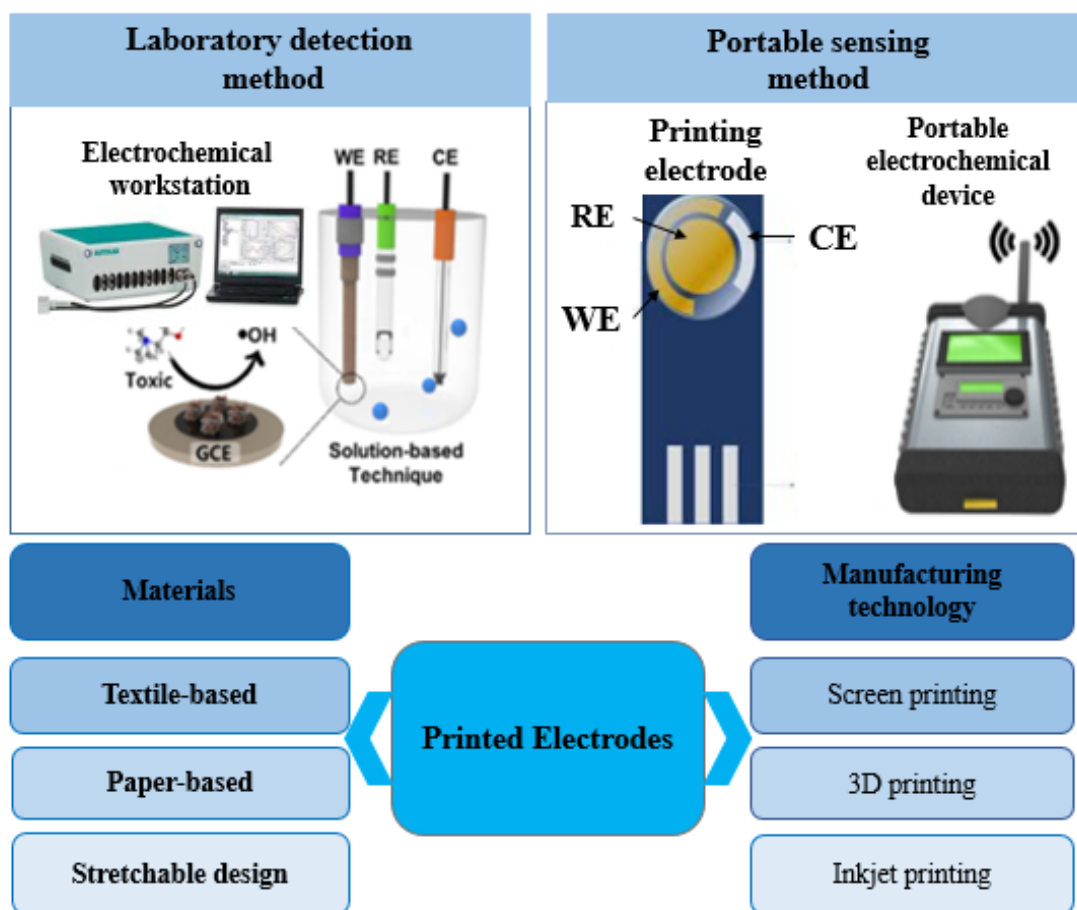


Figure 2. Electrochemical detection methods [13]

García-Miranda Ferrari A. *et al.* in this review explores and discusses the current advances related to electrochemical methods, electrode materials and modifications that have the potential to be the foundations of a new generation of portable electrochemical sensors towards trace-level ion in situ heavy metal sensors. The three-electrode system, as shown in Figure 2, can be placed in a single solution, which in turn allows the device to be automated using a small amount of reagent mixtures and a small sample volume to be examined [13, 14]. Methods for electrochemical sensitivity determination are shown schematically in Figure 2, comparing a laboratory scale electrochemical device with a portable electrochemical device, screen printed electrode (SPE) and classical type electrode fabrication methods.

Screen printed electrode (SPE) is considered to be the first type of electrode that has become a good alternative, overcoming the problems and limitations of traditional electrochemical systems and electrodes. Screen printing electrodes are characterized by single-use, simplicity, portability and fast operation. Due to its versatility and wide use, SPE is widely used as a platform for electrochemical methods for real-time determination of food, gases, pesticides, pharmaceutical additives, environmental contaminants and biomolecules. It should be noted that electrodes fabricated by screen printing technology are generally manufactured in a wide range because the manufacturing process is automated and economically designed. Inkjet printing can be made of plastic or ceramic substrates and different types of inks (usually graphite, carbon, gold and silver) are used for printing. In recent years, the inks used to improve the chemical, electrochemical, analytical and electroanalytical properties of sensors and biosensors based on the above method are modified with various enzymes or nanomaterials. Inkjet printing is created using computer

software and grouting method which results in a negative image of the intended electrode geometry [15-17]. Table 1 summarizes the comparative advantages and disadvantages of the above types of electrodes [18].

Kaur *et al.* [1] demonstrated the use of biomolecules, three-dimensional (3D) printing and smartphone-based electrochemical biosensors for the detection of organophosphorus compounds.

Table 1. Overview of the advantages and disadvantages of electronic printing technology [18]

	Advantages	Disadvantages
Inkjet printing	<ul style="list-style-type: none"> - Cost-effective, simple and convenient - No need to make a pattern or ink in advance - Less material required - Forms thin layers <1 micron thick - High quality, productivity and speed of gel formation - Ease of mass production - Electronic control of ejection rate and drop size - Thin layers 	<ul style="list-style-type: none"> - Highly viscous bio-inks cannot be use - Conductivity problems - Poor mechanical strength - Low structural reliability - Clogging of nozzle - Difficult to apply ink to the substrate
Screen printing	<ul style="list-style-type: none"> - Possibility to produce different electrodes with high reproducibility - Process simplicity - Portability, versatility, simplicity, low cost, reliability and ease of operation - Thick layers - For small batch laboratory analysis 	<ul style="list-style-type: none"> - Difficult to use on a large scale - Low quality - Separate screen required for each print template - Use of templates, photomasks or other physical props to facilitate template use
3D printing	<ul style="list-style-type: none"> - Computer-aided design - Cost-effective - No need for external electrodes - Can be miniaturized - Enables the creation of complex structures with precise dimensional control - Suitable for on-site analysis - Easy installation or customization - Electrodes can be printed without substrate - Light weight, high flexibility and roughness 	<ul style="list-style-type: none"> - Low quality - Limited quantity of raw materials - Application surface cannot be accessible - Difficulty in using materials other than polymer matrices - Difficulty in manufacturing integrated electrodes

Voltammetric detection method. The voltammetric detection method determines the current in the potential difference used by analyzing. The characteristic advantages of voltammetric sensors are high sensitivity to detect and measure several substances simultaneously [19]. The basis of the voltammetric detection method is potential control. There are various methods and forms of potential change such as line scan, polarography (constant voltage), differential ladder, reverse pulse, normal pulse, differential pulse, etc. Among them, cyclic voltammetry, square wave voltammetry, differential pulse voltammetry and electrochemical impedance spectroscopy are the most commonly used methods for

electrochemical detection of pesticide residues in food.

Cyclic voltammetry (CV) is an electroanalytical method that is mainly used to measure electroactive redox forms by linearly varying the potential between the working electrode and the reference electrode. In cyclic voltammetry, the peak current is directly proportional to the concentration of the substance, and the position of the peak is mainly dependent on the chemical and physical species involved in the redox reactions. When scanned, the chemical species either gains an electron (reduction) or loses an electron (oxidation) depending on the direction of change in potential. The voltage is scanned between the working electrode and the reference electrode, and the current is tested between the opposite electrode and the working electrode. The resulting electrochemical analysis can be referred to as current-voltage relationship or voltammetry method. For example, an increase in the concentration of certain enzymes interacting at a given scan rate results in an increase in current compared to a non-catalytic reaction. The cyclic voltammetry method can be used for local analysis of pesticide residues [11, 12].

Bakytkarim Y. *et al.* [20] used the methods of cyclic voltammetry and differential pulse voltammetry to study and compare the electrochemical behaviour of chlorogenic acid on electrodes with different modifications. Figure 4A shows the CV plots of $20\mu\text{mol dm}^{-3}$ chlorogenic acid on bare GCE and Pt@r-GO@MWCNTs/GCE-containing in 0.1 M PBS buffer solution at pH 6.0. As can be seen, when bare GCE was the working electrode, a pair of redox peaks appeared around 0.21 V, which indicated that GCE had some catalytic effect on the oxidation and reduction of chlorogenic acid. When the Pt@r-GO@MWCNTs nanocomposites were modified onto GCE, we observed a large back current and the electrochemical signal of chlorogenic acid was enhanced. As can be seen in Figure 4B, compared with bare GCE, chlorogenic acid showed a very obvious oxidation peak on Pt@r-GO@MWCNTs/GCE, and the current signal of the oxidation peak was amplified nearly 60 times. This is attributed to the relatively large specific surface area, good electrical conductivity and catalytic properties of the Pt@r-GO@MWCNTs nanocomposites. Therefore, Pt@r-GO@MWCNTs/GCE can be used for the detection of chlorogenic acid.

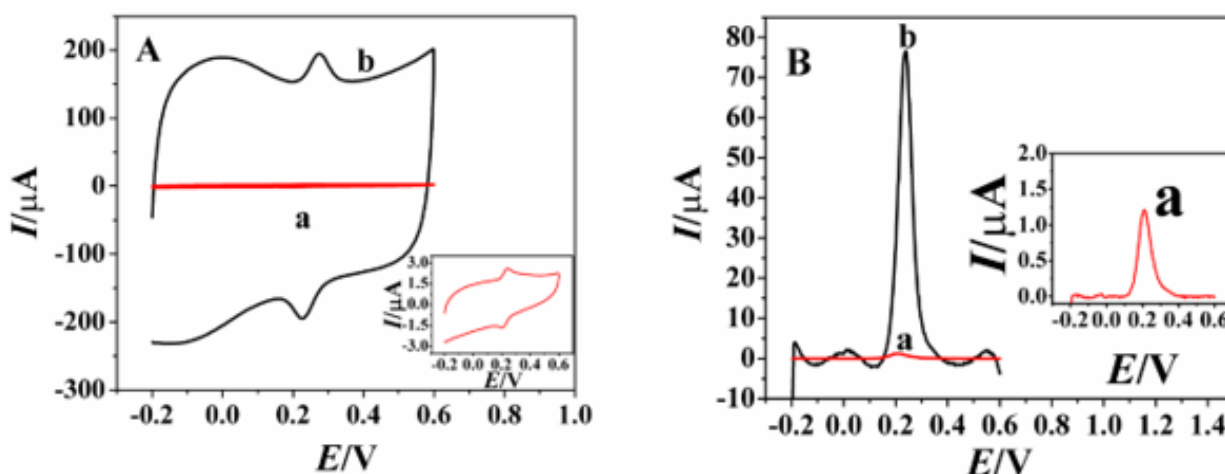


Figure 4. (A, B) Cyclic voltammograms of $20\mu\text{mol dm}^{-3}$ chlorogenic acid in 0.1 M PBS (pH = 6.0) buffer on bare GCE and Pt@r-GO@MWCNTs/GCE Differential pulse voltammograms [20]

Zhao *et al.* [21] developed a sensor based on the retardation of AchE enzyme activity. This sensor often monitored the thiocholine oxidation current through a three-electrode potentiostat with its structure. For pesticide analysis, AchE/MWNTs-SnO₂-chitosan/Au was immersed in phosphate-buffered saline (pH 7.5) with different concentrations of pesticide

solution for 10 min and then transferred to an electrochemical laboratory. The sensor developed signal processing and detection modules to reduce system drift and interference as it emitted low current signals. The value of the obtained detection threshold for chlorpyrifos was 2 mg/l, which indicates the high stability and accuracy of the device. This system proved its efficiency in the determination of pesticide residues in fruit and vegetable samples. To validate the developed portable device, the current signal responses for chlorpyrifos were compared with the electrochemical device at different concentrations. As a result, the authors proposed a portable instrument developed for rapid monitoring of pesticide residues in fruits and vegetables, which was economical and characterised by the possibility of use in the field [21].

Differential pulse voltammetry (DPV) is a pulse method for reducing background charge currents. It uses amplitude pulses of linear potential and the basis of this method is the value of the basic potential, which is not a Faraday reaction in an electrochemical system. The basic potential value is increased equally and in equal increments between pulses. In this case, the current is measured before the pulse is transmitted and the difference between the pulses is noted at the end of the pulse. Differential pulse voltammetry is also used for chemical quantitative analysis as well as for studying the mechanism, thermodynamics and kinetics of chemical reactions. This method as an analytical tool has several advantages over other electrochemical methods. This method as an analytical tool has several advantages over other electrochemical methods. It is a sensitive method that often allows direct analysis at the ppb (10^{-7}). When the voltage attenuation mode is used, ppt (10^{-8}) can be measured. The sensitivity of this method depends on differential analysis and relatively short pulse times. Differential analysis allows the differentiation of background procedures, and the relatively short pulse time amplifies the measured currents [2, 11, 12].

Electrochemical impedance spectroscopy (EIS) is an efficient detection method used to study electrolyte/electrode surface modification and analyze the interactions between bioreceptor and analyte on the electrode surface by observing the changes in the surface resistance of the solution/electrode. In the past few years, the EIS method has been widely used in the field of electrochemistry, especially for the classification of sensor systems. In addition, EIS is also used as an analytical and quantitative determination method. The EIS method can evaluate the intrinsic material properties that affect the resistance, conductivity or capacitance of an electrochemical system. Thus, the EIS method is an effective tool for the design and development of materials for sensors and biosensors [19].

As most sensing systems are inherently heterogeneous at the nanoscale, with their variability in the interfacial region of the sensing electrodes heterogeneity may not be apparent using pulse voltammetric methods, square wave voltammetry or cyclic voltammetry. The EIS method makes it easy to study this problem as well as see it through a wide range of time scales. The sensors and biosensors underlying the EIS method have great advantages over other modern signal transducers used to detect pesticide residues.

Square wave voltammetry (SWV) is the most sensitive method of differential and fast pulse voltammetry with large amplitude. The detection limit values of real samples obtained by square wave voltammetry can be relatively better than spectroscopic and chromatographic methods. The values of the current potential peaks are usually well defined and present symmetrical plots. This method is due to the fact that all currents are investigated only at the end of a single half-period and to differences in the width and height of the potential pulse. Square wave voltammetry is a sensitive electrochemical detection method used for recognition on paper or integrated devices based on the signal-to-noise phenomenon. An increase in the square root of the scan rate results in an increase in the signal-to-noise ratio. This increase in signal is a function of the time between current measurement and pulse transmission. The method-time is frequency-dependent; increasing the frequency results in a

decrease in the time constant and an increase in the measured Faraday current, which in turn makes it possible to quickly display specific pulses on the voltammetry. There will be fast delivery of the obtained voltammetry results to the consumer via smartphone. Recent advances in the creation of the material that will form the basis for sensors such as flexible, portable and elastic electronic devices are gradually improving the quality of human life.

Today's wearable sensors can be attached directly to the user's body or to the surface of an object to perform real-time laboratory and chemical analyses. Inspired by this approach, Zhao et al. In situ developed a smart hand-worn plant sensor that enables fast, sensitive and non-destructive detection of pesticide residues on plant surfaces [22]. A serpentine three-electrode system was designed by laser-induced graphene technology. This handheld portable electrochemical system can wirelessly deliver real-time info about pesticide residue to a smartphone connected to the handheld workstation via Bluetooth. Pesticides were determined between 0.6 V and + 0.4 V in a solution of different concentrations using SWV analysis method at a frequency of 8 Hz with an equilibrium time of 30 seconds.

Spinach and apples were selected as specific samples for in situ methyl parathion analysis. Using the developed approach, 100 μ M of methyl parathion was detected in agricultural samples. Figure 4 (a) shows a digital photograph of elongated and flexible polydimethylsiloxane laser-induced graphene (PDMS LIG). SEM images of PDMS/LIG electrodes 7 (b) and (c) are presented before and after modification with gold nanoparticles (AuNPs). Inset (c) shows a magnified image of the SEM after modification with AuNPs. Figure 4 (d) and (g) compared to control experiments, p-nitrophenol peaks were observed in samples coated with methyl parathion indicating the presence of pesticide residues at a specific location. The smart wearable sensor device demonstrates advances and transformative approaches to in situ pesticide residue analysis in agricultural products. The system can be used on uneven surfaces that selectively recognise and capture pesticide residues on crop surfaces [23].

The information of pesticide residues as shown in Figure 1 can be obtained via smartphone at the same time. Compared to control experiments, peaks of p-nitrophenol were observed in the samples coated with methyl parathion, indicating the presence of pesticide residues at a specific location. A smart, lightweight, wearable sensor device demonstrates progress and innovative approaches to in situ analysis of pesticide residues in agricultural foods. Such devices can be used to selectively detect pesticide residues on the surface of plants located on uneven surfaces.

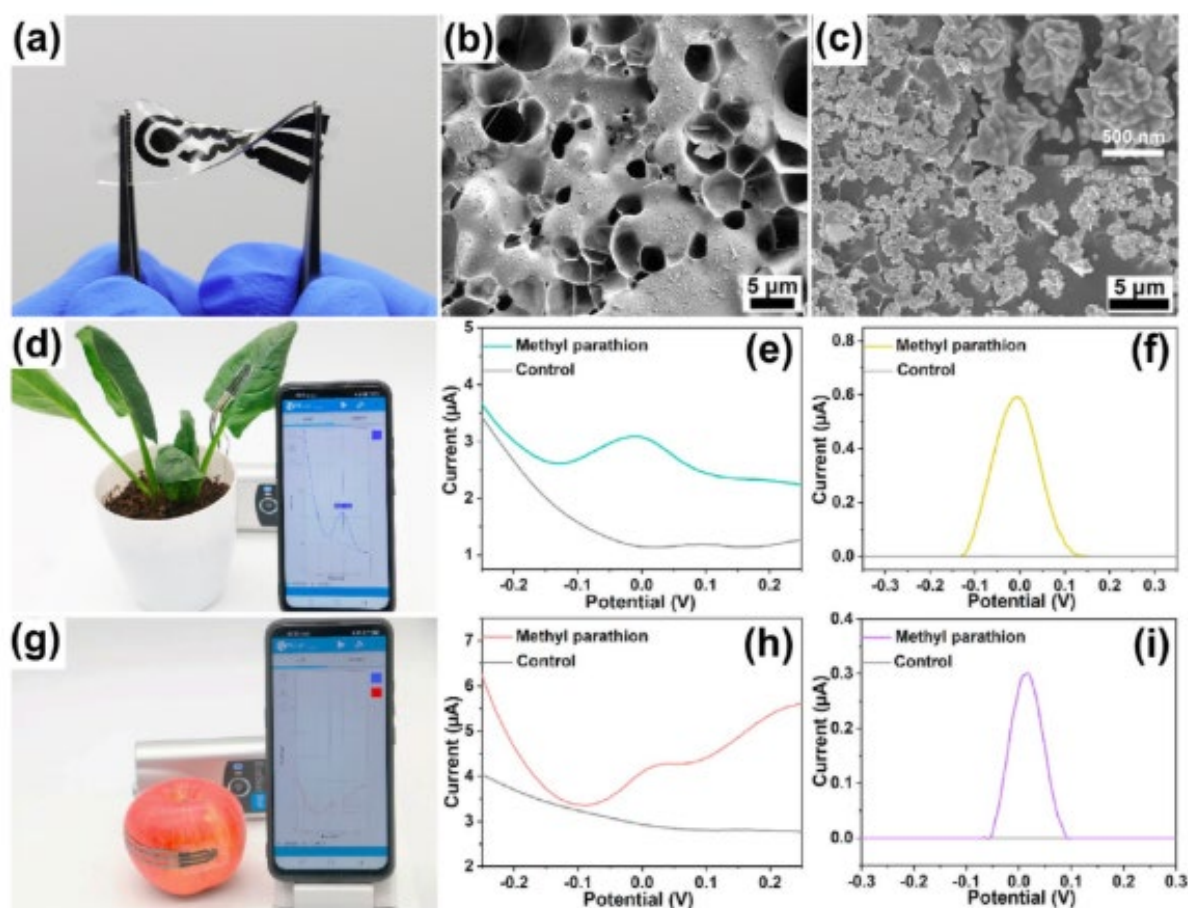


Figure 4. *a* - Digital photograph of the stretchable and flexible (PDMS-LIG) electrode, *b, c* - SEM images of the electrode before and after modification with gold nanoparticles (PDMS-LIG) (AuNPs), enlarged SEM image after modification *c* - AuNPs. *d, g* - flexible semi-rigid apple/spinach electrolyte biosensor timely information on pesticide residues obtained by smartphone adhered to the leaf surface. *e, h* - square wave voltammetry curves and corresponding *f, i*-base corrected square wave voltammetry curves [22]

Conclusion

This review article focuses on electrochemical methods for the investigation of pesticide residues in fruits and vegetables based on cyclic voltammetry, square-wave voltammetry, differential pulse voltammetry, potentiometry and electrochemical impedance spectroscopy. It has been recorded that nano- and microelectrode array chips, paper sensors, paper strips and electroanalytical devices have been developed for rapid and high-throughput multiplexed investigation of complex matrices. Many attempts have been made to develop portable and user-friendly printed electrode devices, cost-effective integrated microsensors, glove-attached sensors, paper/strip/card-based electrochemical sensing platforms. Electroanalytical and electrochemical processes have been significantly improved for in situ testing and detection of pesticides in agricultural products.

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Электрохимиялық әдістерді қолдана отырып жемістер мен көкөністер құрамындағы пестицид қалдықтарын жылдам анықтау

Андатпа. Әлемдік сұранысты қамтасыз ету үшін пестицидтер ауылшаруашылық өнімдеріне зиян келтіретін зиянкестермен күресуде өте маңызды роль атқарады. Алайда пестицидтерді шамадан тыс қолдану экожүйелер мен адам денсаулығына қауіп төндіруде. Соңғы жылдары пестицидтерді анықтауда Газ хроматографиясы, сұйық хроматография, газ хроматографиясы-масса-спектрометрия, сұйық хроматография-масса-спектрометрия, капиллярлық хроматография және капиллярлық электрофорез қолданылады. Бұл әдістердің пестицидтерді анықтау үшін ерекшелігі, селективтілігі және сезімталдығы жоғары болғанымен, олардың кейбір кемшіліктері де бар, мысалы, қымбат құрал, сынама дайындау процесі күрделілігі және көп уақытты қажет етеді. Бұл мақалада жемістер мен көкөністердегі пестицидтердің қалдықтарын орнында анықтауға арналған портативті электрохимиялық сенсорлар жасау әдістері, яғни циклдік вольтамметрия, квадрат-толқындық вольтамметрия, дифференциалды импульстік вольтамметрия және электрохимиялық импеданс спектроскопиясының соңғы жылдардағы жетістіктері туралы баяндалған. Сонымен қатар, көрсетілген әдістердің негізгі механизмі мен электрохимиялық сезгіштігі туралы толық ақпарат берілген. Электрохимиялық әдістердің дамуы және смартфон технологиясын нақты уақыт режимінде бақылау, сонымен бірге оларды анықтау платформаларымен интеграциялау талқыланады. Ғылыми және технологиялық бағыттағы оқырмандар үшін бұл мақала, портативті электрохимиялық құрылғыларды жасауды, пестицидтерді жылдам анықтауды, электрохимиялық сезгіш әдістерінің рөлін түсіну және олардың әрі қарай дамуына ықпал ету үшін қосымша құнды ақпарат болады деген ойдамыз.

Түйін сөздер: пестицидтер, электрохимиялық әдістер, сенсорлар, нанобөлшектер, модификация, смартфон арқылы мониторинг, электрохимиялық құрылғылар.

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Быстрое обнаружение остатков пестицидов во фруктах и овощах с помощью электрохимических методов

Аннотация. Для удовлетворения мирового спроса пестициды играют очень важную роль в борьбе с вредителями, наносящими ущерб сельскохозяйственной продукции. Однако чрезмерное использование пестицидов угрожает экосистемам и здоровью людей. В последние годы для определения пестицидов используются газовая хроматография (ГХ), жидкостная хроматография (ЖХ), газовая хроматография-масс-спектрометрия (ГХ-МС), жидкостная хроматография-масс-спектрометрия (ЖХ-МС), капиллярная хроматография (КХ) и капиллярный электрофорез (КЭ). Хотя эти методы обладают высокой специфичностью, селективностью и чувствительностью для обнаружения пестицидов, они также имеют некоторые недостатки, например, дорогостоящий прибор, сложный и трудоемкий процесс подготовки проб. Поэтому электрохимические сенсоры и биосенсоры стали хорошими аналитическими методами для обнаружения пестицидов благодаря их многочисленным преимуществам, таким как простота изготовления системы обнаружения, высокая чувствительность и селективность. В данной статье описаны последние результаты, полученные в области циклической вольтамперометрии, квадратно-волновой вольтамперометрии, дифференциальной импульсной вольтамперометрии и электрохимической импедансной спектроскопии, используемые для *in situ* обнаружения остатков пестицидов в овощах и фруктах. Кроме того, обсуждаются вопросы развития электрохимических методов и технологий мониторинга с помощью смартфонов в режиме реального времени, а также их интеграции с платформами обнаружения. Читателям с научно-технической направленностью эта статья предоставит дополнительную ценную информацию для понимания создания портативных электрохимических устройств, быстрого обнаружения пестицидов, роли электрохимических чувствительных методов и внесения вклада в их дальнейшее развитие.

Ключевые слова: пестициды, электрохимические методы, сенсоры, наночастицы, модификация, мониторинг с помощью смартфона, электрохимические устройства.

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