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Original scientific paper

ANALYSIS OF ASBESTOS IN BUILDING MATERIALS – CASE IN TOWN SKOPJE

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Asbestos has been extensively researched as one of the most hazardous materials to human health in the past century. Despite its universal recognition as a challenging problem, the topic is unfortunately not widely discussed in our country. However, the city authority of Skopje has undertaken a significant campaign focused on identifying and removing asbestos-containing materials from various buildings in the city. This initiative represents the largest effort of its kind in the country thus far. The campaign involved the development of methodologies and the analysis of samples collected from different buildings in Skopje. A systematic analysis was conducted on 50 samples collected from 21 public buildings in Skopje, utilizing techniques such as optical microscopy, infrared spectroscopy, X-ray diffraction, and scanning electron microscopy (SEM). The results revealed the presence of asbestos in 33 of the analyzed samples. Notably, asbestos was not detected in only two buildings. These findings indicate that asbestos is present in the building materials of practically all the inspected structures, suggesting its likely presence in many other buildings within the city and throughout the country. Of particular concern is the confirmed presence of asbestos in kindergartens, as this exposes the young population to potential asbestos-related health risks.

Key words: asbestos; serpentine asbestos; amphiboles asbestos; building materials; Skopje

INTRODUCTION

Asbestos

The contemporary era of extensive industrialization and globalization has given rise to numerous processes that have severe detrimental effects on human health. The urbanization that followed the industrial revolution inevitably led to the development of various building materials with different origins. However, in the rapid pace of urbanization, many crucial aspects closely tied to long-term human well-being were not adequately anticipated. One prominent example of such prolonged adverse effects is the utilization of asbestos in building materials over an extended period. The health issues associated with the widespread use of asbestos-containing materials have not been fully resolved and continue to be a significant topic of concern in many urban areas.

It is crucial to emphasize that asbestos has been extensively studied as one of the most hazardous materials to human health in the past century. The scientific and medical literature is replete with hundreds of papers addressing various aspects and questions related to asbestos. Additionally, scientific conferences and debates are held annually, focusing on different facets of asbestos use and associated risks [1–19]. Regrettably, in our country, this topic is not extensively discussed, despite its universal recognition as one of the most challenging issues that require attention. The campaign initiated by the municipal authorities of Skopje, aimed at identifying and eliminating asbestos-containing materials from various structures in the city, represents the most significant effort of its kind in the country thus far. This initiative involved the implementation of specific methodologies and the presentation of results obtained from the analysis of samples collected from different buildings within the city.

The term "asbestos" encompasses multiple fibrous silicate minerals that can be categorized into two groups: ***serpentine asbestos*** and ***amphibole asbestos***. The serpentine group primarily includes ***chrysotile***, commonly known as white asbestos, which is the most prevalent form of asbestos. The amphibole group consists of ***amosite*** (brown or grey asbestos), ***crocidolite*** (blue asbestos), ***tremolite, actinolite***, and ***anthophyllite***. Among these, chrysotile has been the most economically significant, accounting for approximately 95 % of the global asbestos production until about two decades ago. Crocidolite and amosite were the predominantly produced types within the amphibole asbestos category, while anthophyllite, tremolite, and actinolite were produced to a lesser extent. It is worth noting that the use of any asbestos-containing products was completely banned in European Union countries in 2005.

Asbestos minerals possess several commercially valuable properties, including high strength, elasticity, thermal resistance, and chemical stability. These characteristics make asbestos particularly suitable for manufacturing textile products and materials that need to withstand heat, fire, and friction. Not all naturally occurring fibrous minerals are classified as asbestos since they do not possess these properties. Asbestos is typically not used or found in its pure form; it is often mixed with other materials. Consequently, visual inspection alone is not conclusive for identifying asbestos due to its stable nature and infrequent occurrence in its native form.

Serpentine minerals have emerged as a captivating subject of research [20, 21] due to several compelling reasons.

– Firstly, they represent the last group of minerals in rocks for which the detailed structural polytypes have not been clearly described.

– Secondly, there is scientific and practical interest in understanding the thermodynamics of their formation.

– Lastly, their significant impact on the living and working environment has drawn considerable attention.

The heightened concern surrounding asbestos, a mineral known to pose serious health risks, has substantially increased interest in its study and contributed to advancements in the field of mineralogy. When materials containing asbestos are damaged or broken, they can release asbestos fibers into the air, which can be inhaled and lead to adverse health effects. Prolonged exposure to airborne asbestos has been linked to several negative health outcomes, including asbestosis, lung cancer, and mesothelioma [3–6, 9, 12, 14, 16, 17, 19].

Despite the abundance of available data on these diseases, scientists still lack a comprehensive understanding of the underlying mechanisms by which asbestos triggers them. It is worth noting that inhaling *amphibole asbestos* has been proven to pose a significantly higher health hazard compared to inhaling *serpentine asbestos* [3, 12].

Detection of materials containing asbestos

Materials that contain asbestos (referred to as asbestos materials, AM) are classified by the U.S. Occupational Safety and Health Administration (OSHA) and the U.S. Environmental Protection Agency (EPA) as "materials containing more than one percent (1%) asbestos".

Thermal insulation systems (TSI) and flooring materials installed prior to 1980 are highly likely to contain asbestos. Additionally, asphalt and vinyl flooring materials produced before 1980 are also suspected to contain asbestos. Therefore, it is essential to inspect these materials to confirm or eliminate any doubts regarding the presence of asbestos.

When materials are suspected to contain asbestos, it is necessary to conduct thorough checks to confirm its presence. If asbestos is confirmed, the owners of buildings, structures, and equipment are obligated to inform residents and contractors about the presence, location, and asbestos content in their properties. If asbestos-containing materials are damaged or deteriorated, they can release fibers into the surrounding air within the living or working environment, posing a potential risk of inhalation and subsequent health issues. It is important to note that intact asbestos-containing materials do not pose a health hazard.

Review of asbestos regulation in the European Union (EU)

Asbestos and materials containing asbestos are subject to regulation under several EU directives, including the following:

- Regulation (EC) No 1907/2006, known as the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) Regulation. This directive, enacted on 18 December 2006, establishes the European Chemicals Agency and amends various previous directives. It repeals Council Regulation (EEC) No 793/93 and Commission Regulation (EC) No 1488/94, as well as Council Directive 76/769/EEC and Commission Directives 91/155/EEC, 93/67/EEC, 93/105/EC, and 2000/21/EC.

Annex XVII of the REACH Regulation (EC) No. 1097/2006, which bans the marketing and use of products containing asbestos in the European Union. This regulation prohibits the use of asbestos in new products.

- Regulation (EC) No 1272/2008, enacted on 16 December 2008, concerning the classification, labelling, and packaging of substances and mixtures. It amends and repeals Directives 67/548/EEC and 1999/45/EC and also amends Regulation (EC) No 1907/2006.
- Directive 2009/148/EC, enacted on 30 November 2009, of the European Parliament and the Council of Europe. This directive aims to protect workers from the risks associated with asbestos exposure in the workplace. It establishes limit values for exposure to asbestos and sets forth specific requirements to prevent and mitigate health risks related to asbestos exposure at work.

The aforementioned directive focuses on safeguarding workers' health and aims to prevent risks arising from exposure to asbestos. It sets limits for acceptable exposure levels and outlines various specific requirements. The most significant requirements are detailed below:

- **Prohibition** of the application of asbestos through spraying processes and working procedures involving the use of low-density insulating or soundproofing materials (with a density less than 1 g/cm^3) that contain asbestos.

- **Prohibition** of activities that expose workers to asbestos fibers during asbestos extraction, manufacturing and processing of asbestos products, and manufacturing and processing of products intentionally containing asbestos (excluding products resulting from demolition and asbestos removal).

- **Employers** must ensure that workers are not exposed to an airborne concentration of asbestos exceeding 0.1 fibers per 1 cm^3 as an 8-hour time-weighted average.

- Regular measurement of asbestos fibers in the air at the workplace should be conducted, depending on the results of the initial risk assessment, to ensure compliance with the aforementioned limit value.

- Commission Regulation (EU) 2016/1005 of 22 June 2016, amending Annex XVII to Regulation (EC) No 1907/2006, focuses on asbestos fibers (chrysotile).

Furthermore, EU members emphasize the need for an impact assessment and cost-benefit analysis to consider the establishment of action plans for the safe removal of asbestos from public buildings and buildings with regular public access by 2028. They also urge the provision of information and guidelines to encourage private homeowners to conduct effective audits and risk assessments of their premises for asbestos-containing materials (ACMs), following the example of Poland.

In this context, it is important to highlight the statement of the International Commission on Occupational Health (ICOH) advocating for a global ban on the production, sale, and use of all forms of asbestos, as well as the elimination of asbestos-related diseases.

“The ICOH calls on each country to implement a total ban on asbestos production and use, and to pursue primary, secondary, and tertiary prevention of asbestos-related diseases through country-specific "National Programs for the Elimination of Asbestos-Related Diseases," following guidelines from the International Labor Organization (ILO) and World Health Organization (WHO).”

The current regulatory framework for asbestos in Macedonia, which aims to align with EU standards, is not yet fully completed. The existing regulations pertaining to asbestos treatment are as follows:

- The maximum permissible concentration of asbestos in the air emitted from sources is 0.5 mg/m^3 per hour, and the maximum allowable quantity of asbestos in the air is 0.1 mg/m3 per hour (Official Gazette of RM No. 3 in 1990).
- The presence of asbestos in water is regulated by the Water Classification Regulation (Official Gazette of RM No. 18 in 1999), which prohibits the presence of asbestos in any of the IV classes of water.
- Guidelines for handling waste containing asbestos and asbestos materials are outlined in the Law on Waste Management (Official Gazette of RM No. 68/04 and 71/04 in 2004).
- The Rulebook on minimum requirements for workers' health and safety concerning risks related to asbestos exposure at work (Official Gazette of RM No. 50 in 2009) provides specific regulations in this regard.

ACTIVITIES

Considering the aforementioned health hazards associated with asbestos and the timeframe for their manifestation (5–40 years after initial exposure to airborne asbestos), it is crucial to conduct thorough inspections of all public buildings in the country, particularly those predominantly occupied by the young population who will enter their most productive years in 15–20 years' time. Given the significant risks posed by asbestos to human health and well-being, especially among the young population, it is urgent to establish a schedule for identifying the presence of asbestos materials and their removal from all public institutions in Macedonia.

In this regard, inspections and analyses have been conducted in numerous public institutions in the city of Skopje, including kindergartens and schools, and the results are presented in this article. These findings provide valuable data regarding the incorporation of asbestos-containing materials in public buildings, which necessitates their proper treatment and elimination to mitigate potential health risks associated with asbestos exposure.

EXPERIMENTAL

The collected material was prepared for optical investigation using an optical binocular and the transmission of polarized light. This method only provides an indication of the presence of asbestos in the examined material. Infrared spectroscopy was also utilized as an additional technique to study the collected materials. To confirm the presence of asbestos, X-ray diffraction and scanning electron microscopy (SEM) were employed. There is existing literature on the use of these methods for asbestos analysis in various materials [7, 22–29].

The infrared spectra were recorded using a Perkin-Elmer System 2000 infrared interferometer, with pressed KBr disks, at room temperature (RT). To improve the signal-to-noise ratio, 32 scans were collected and averaged. The instrument had a resolution of 4 cm–1 for all measurements. Spectra acquisition and management were performed using the GRAMS ANALYST 2000 [30] and GRAMS 32 [31] software packages.

The presence of asbestos was initially assessed using optical microscopy after appropriate sample preparation. The AX-IOLAB Carl Zeis Jena microscope was used for this purpose.

To identify the studied samples, X-ray diffractograms were recorded and analyzed using the SIMENS D 500 system, controlled by a computer. The analysis was conducted with CuKα monochromatic radiation at 40 kV/30 mA. The computer PDP 11/23+ automatically obtained the optimal signal-to-noise ratio. The characterization of the identified phases was performed using the DIFRAC 11 software package, specifically the EVAL and IDR programs. Samples with minimal orientation were prepared to record an adequate diffraction range of $2\theta = 3\text{-}70\ ^{0}$.

RESULTS AND DISCUSSION

Preliminary monitoring was conducted to detect the presence of asbestos in building materials obtained from various educational and healthcare institutions in Skopje. Multiple samples were collected from different parts and materials of each object, including floors, roofs, walls, and ceilings. In total, fifty samples were analyzed using four different analytical methods: optical microscopy with polarized light, X-ray diffraction, scanning electron microscopy, and infrared spectroscopy.

The results of all the analyses are presented in Table 1, and supplementary material (Figures 1-9) is provided, including infrared spectra, X-ray diffractograms, and SEM images of representative samples containing orthochrysotile, anthophyllite, and both anthophyllite and clinochrysotile. The names and addresses of the public buildings are not disclosed here but can be made available to the relevant authorities in Skopje.

After a systematic analysis of the fifty samples from twenty-one public buildings in Skopje using optical microscopy, infrared spectroscopy, X-ray diffraction, and SEM, it can be concluded **that asbestos is present in 33 of the analyzed samples**. Notably, out of these thirty-three samples with asbestos, twenty-eight contained the serpentine type (chrysotile), while five samples contained the more hazardous amphibole type (anthophyllite), and two samples contained both types of asbestos. It is important to highlight **that asbestos was not detected in only two objects**. These findings suggest that asbestos is present in the building materials of nearly all inspected objects, indicating its likely presence in numerous other buildings throughout the city and the country. The confirmed presence of asbestos in kindergartens is particularly concerning due to the exposure risk it poses to young children. Previous studies have also identified asbestos in the building materials of the Pediatric Clinic in Skopje.

Considering the widespread presence of asbestos in Skopje's buildings, the authors of this study recommend immediate action to safely remove asbestos materials from public buildings. The highest priority should be given to the Pediatric Clinic, kindergartens, primary and secondary schools, and all institutions where children spend a significant amount of time. The second priority should be the removal of asbestos-containing materials from healthcare facilities, followed by sports facilities, and so on.

Table 1. Analysis of asbestos in building materials in different objects in Skopje

No.	**Object**	**Roof**	**Floor**	**Wall**	**Ceiling**
1.	Kindergarten			I/29 S **Nd**[*]	I/29 T **Chrysotile**
2.	Elementary school – 1	I/3a K **Chrysotile**	I/5 n **Nd**		
3.	Elementary school – 2		I/8 n **Nd**	I/8 S **Chrysotile**	I/8 T **Chrysotile and Clinochrysotile**
4.	Elementary school – 3		I/21 n **Chrysotile**	I/21 S **Chrysotile**	I/21 T **Anthophyllite**
5.	Elementary school – 4	I/28 K **Chrysotile**	I/28 n RA 17411 **Chrysotile**	I/28 S **Chrysotile**	I/28 T RA 17388 **Chrysotile (Clinochrysotile)**
6.	Elementary school – 5	I/38 K **Nd**		I/38 S **Chrysotile (Clinochrysotile)**	
7.	High school – 1	I/3 K **Chrysotile (Orthochrysotile)**			
8.	High school – 2	II/3 K **Chrysotile (Clinochrysotile)**			
9.	High school – 3		II/7 n **Chrysotile (Clinochrysotile)**	II/7 S **Chrysotile (Clinochrysotile)**	II/7 T **Anthophyllite**
10.	High school – 4	II/9 K **Nd**	II/9 n **Nd**		
11.	High school – 5	II/13 K **Anthophyllite and Clinochrysotile**	II/13 n **Nd**		
12.	High school – 6	II/14 K **Anthophyllite and Clinochrysotile**	II/14 n **Nd**	II/14 S **Chrysotile (Clinochrysotile)**	II/14 T RA 17425 **Anthophyllite**
13.	High school – 7	II/17 K **Nd**	II/17 n **Nd**	II/17 S **Chrysotile**	II/17 T RA 17428 **Chrysotile**
14.	High school – 8		II/18 n **Nd**		
15.	High school – 9	II/19 K **Nd**	II/19 n **Chrysotile (Clinochrysotile)**	II/19 S **Nd**	
16.	High school – 10	II/20 K **Nd**	II/20 n **Chrysotile**		
17.	Faculty – 1	I/7 K **Chrysotile**	I/7 n **Chrysotile**		
18.	Health care center	I/40 K **Nd**	I/40 n **Nd**	I/40 S **Chrysotile**	
19.	Sports association	I/27 K **Chrysotile**	I/27 n **Nd**		I/27 T **Chrysotile**
20.	House of culture	I/22 K **Chrysotile**			
21.	Public enterprise	I/23 K **Chrysotile (Clinochrysotile)**		I/23 S **Chrysotile**	

*Nd – not detected

When planning the removal of asbestos materials, priority should be given to those containing ***amphibole asbestos*** due to its easier penetration into the human respiratory system. This is primarily because of the dimensions and properties of the fibers in this type of asbestos, as mentioned earlier.

Before initiating the asbestos removal work in any building or structure, it is crucial to conduct a comprehensive inspection and monitoring of the entire premises, following the appropriate guidelines and instructions for asbestos handling and removal. To ensure the safe removal of asbestos-containing materials, it is also essential to differentiate between various types of these materials, such as brittle asbestos, asbestos coatings used for thermal and acoustic insulation in buildings, decorative coatings, asbestos-containing insulation in boilers and other industrial plants, products made of asbestos-containing cement, vinyl-asbestos floors, and various wall coatings.

To facilitate safe removal, **a detailed plan should be prepared for the removal of asbestos-containing materials** from buildings, equipment, factories, and other structures. This plan should include information about the equipment to be used, the techniques for asbestos removal, and the necessary safety and hygiene requirements. It is mandatory to specify a suitable location and method for the safe disposal of asbestos-containing waste. The disposal of asbestos waste must adhere to the regulations outlined in the Rulebook for the methodology of handling waste containing asbestos and waste from asbestos-containing materials, which is based on the Law on Waste Management (Official Gazette of RM No. 68/04 and 71/04 in 2004).

Contractors working with products that may contain asbestos must ensure that all required steps are taken to confirm the presence or absence of asbestos. If there is any doubt about the presence of asbestos, laboratory analysis should be conducted.

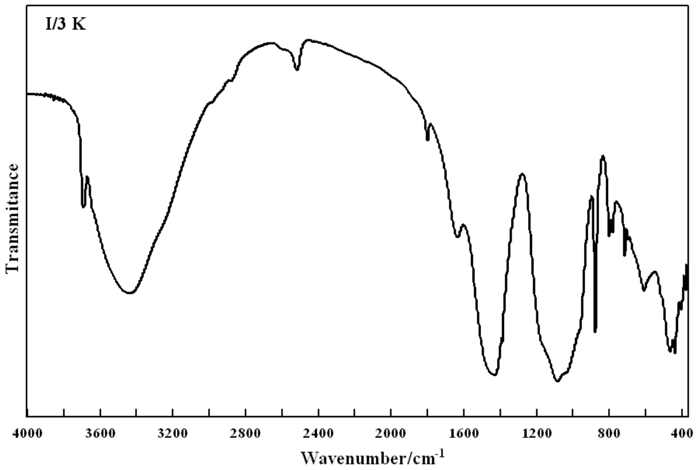

Figure 1. Infrared spectrum of a sample taken from the roof of High school – 1

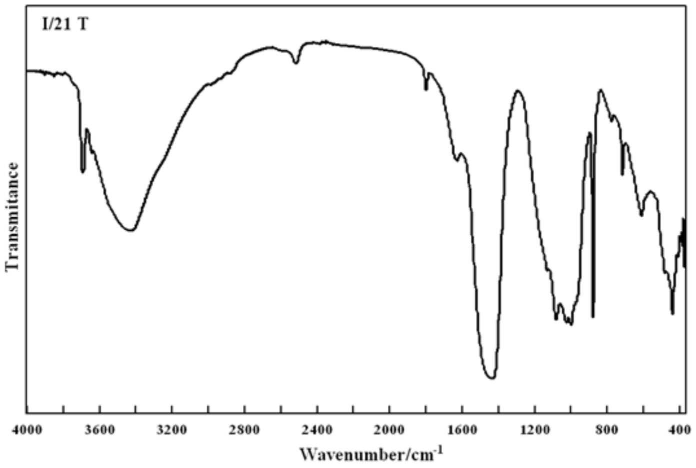

Figure 2. Infrared spectrum of a sample taken from the ceiling of Elementary school – 3

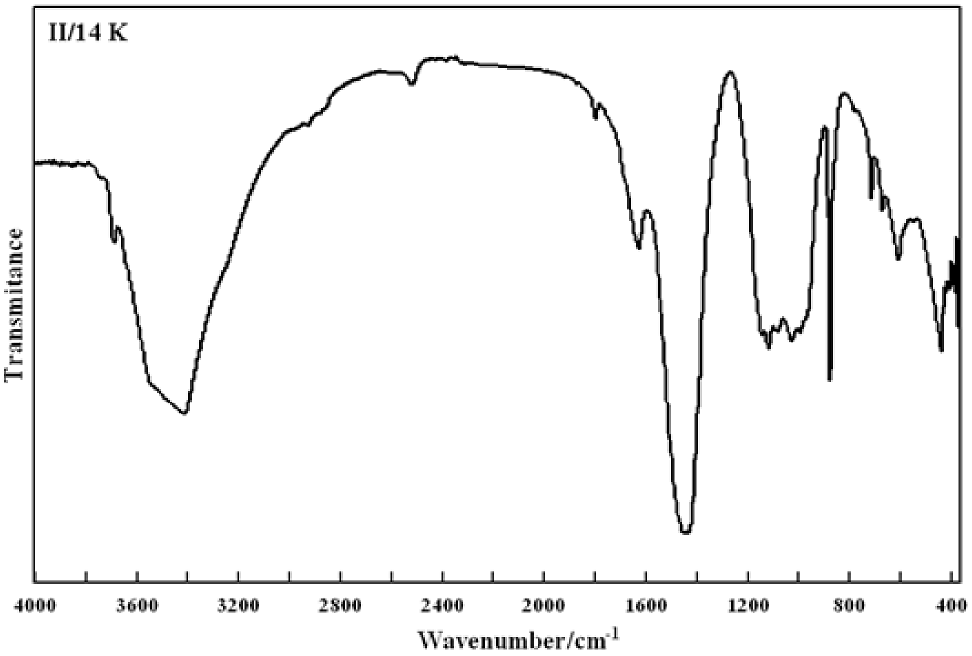

Figure 3. Infrared spectrum of a sample taken from the roof of Elementary school – 6

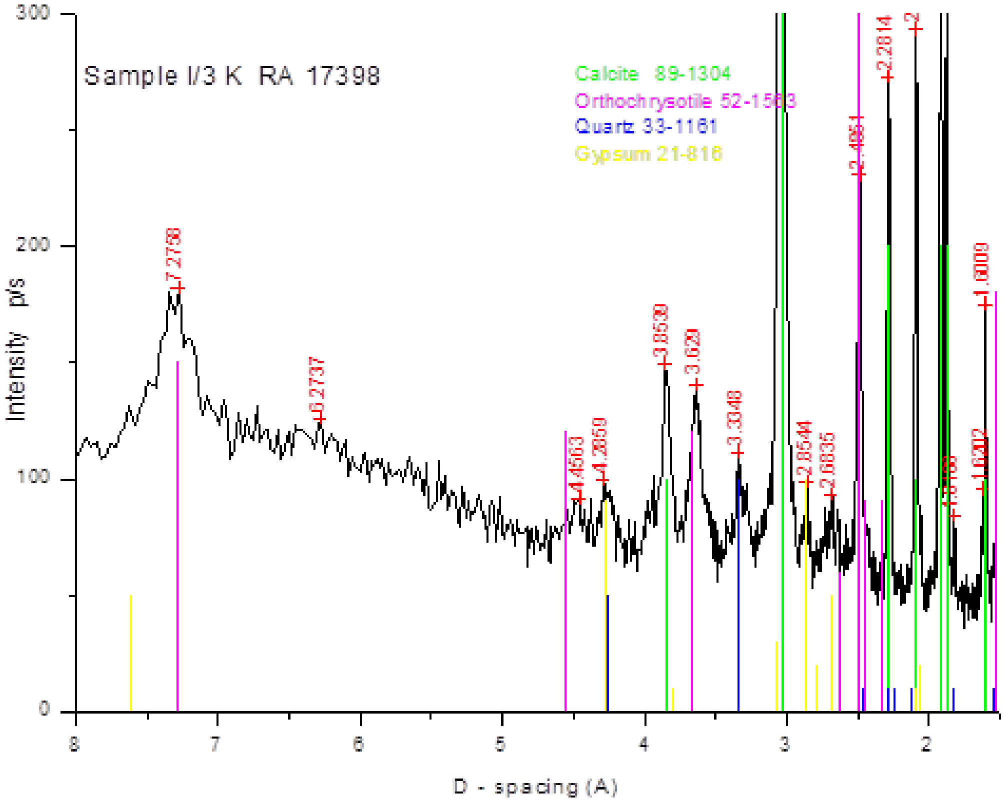

Figure 4. Scattered intensities as a function of the diffraction spacing of a sample taken from the roof of High school – 1

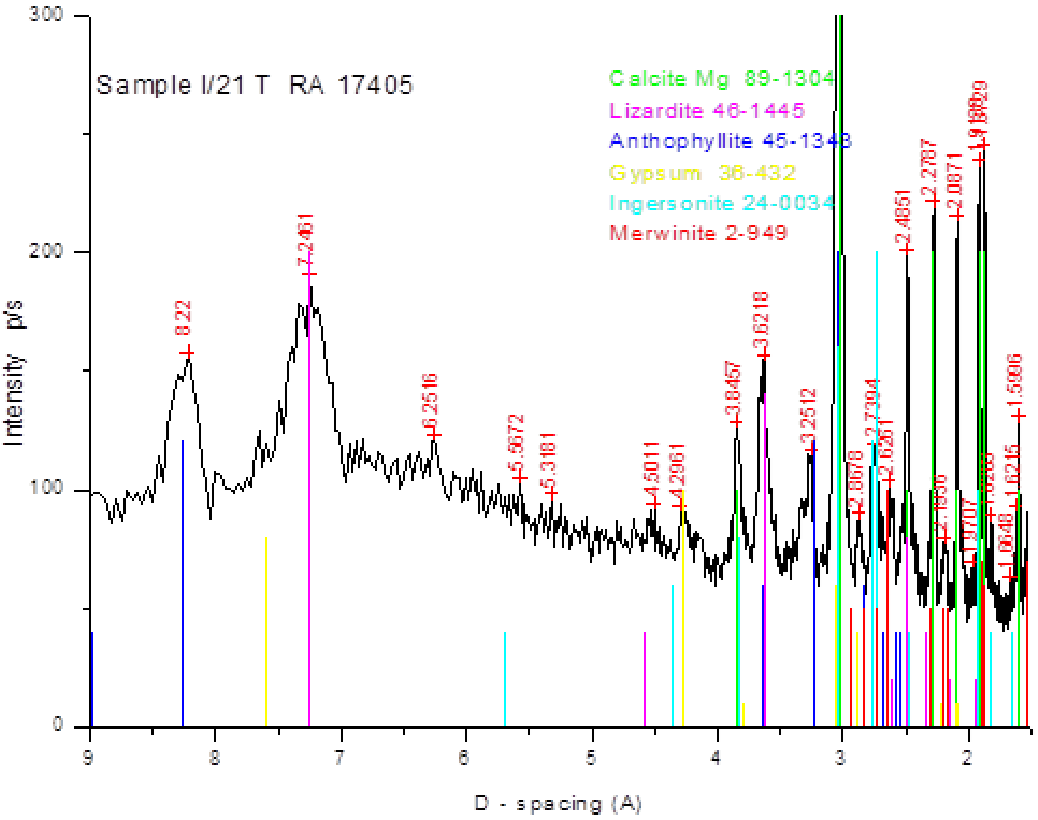

Figure 5. Scattered intensities as a function of the diffraction spacing of a sample taken from the ceiling of Elementary school – 3

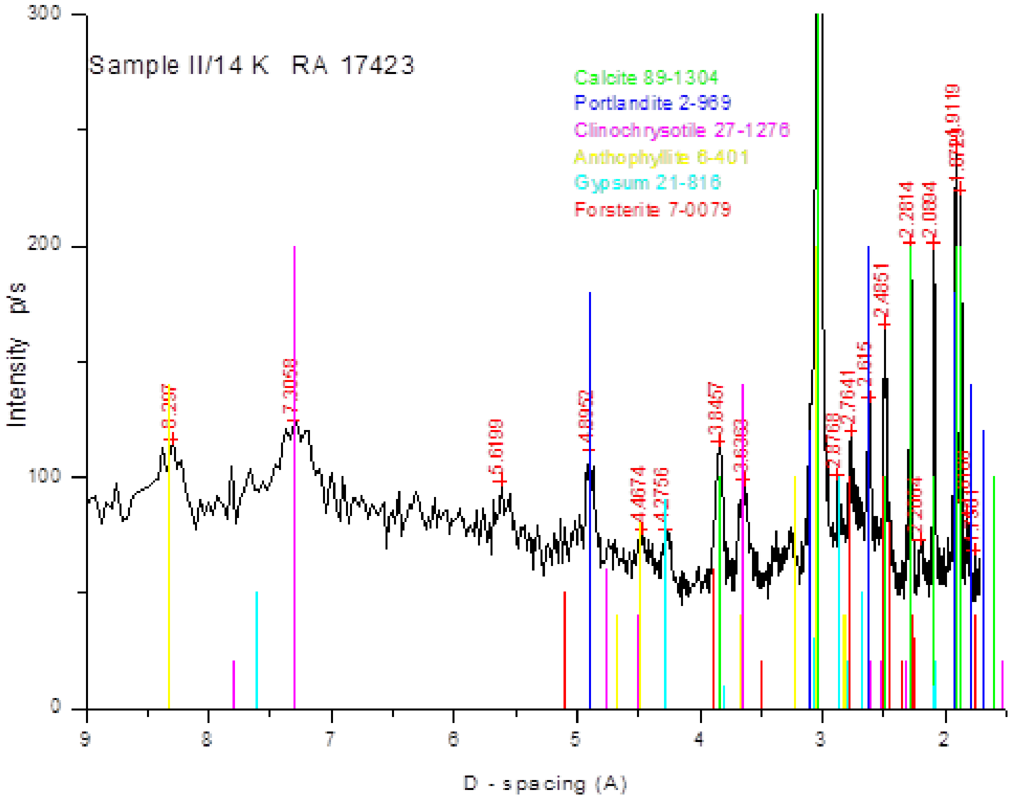

Figure 6. Scattered intensities as a function of the diffraction spacing of a sample taken from the roof of Elementary school – 6

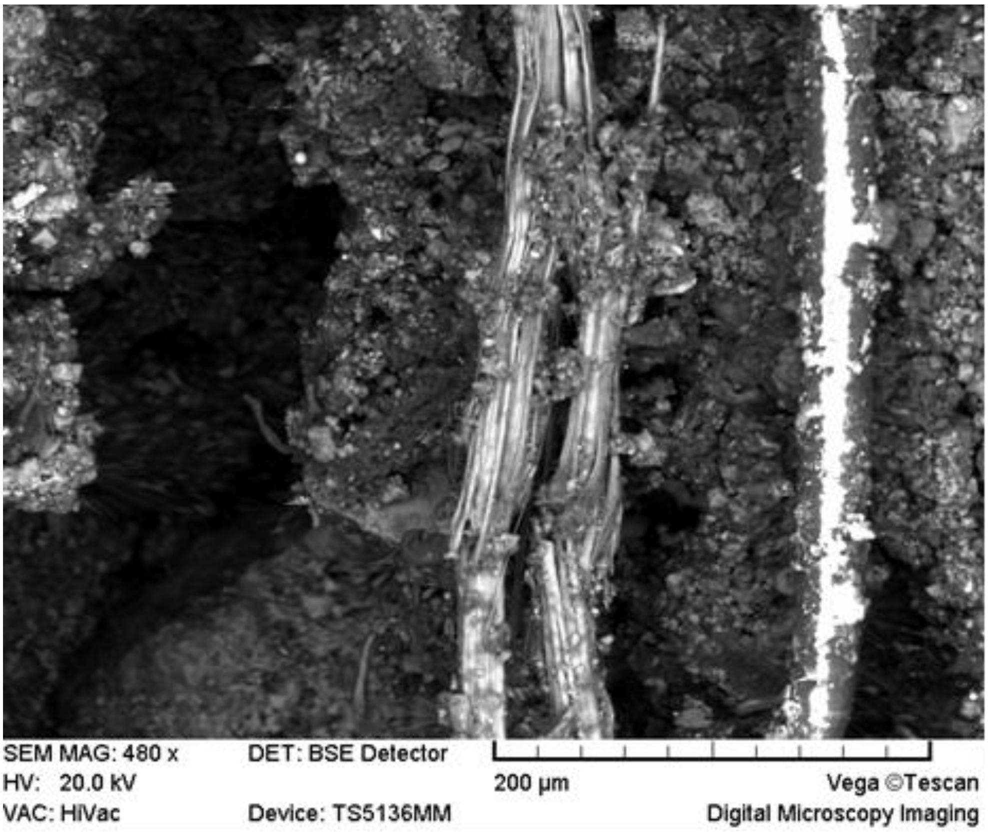

Figure 7. SEM of selected sample that contain asbestos

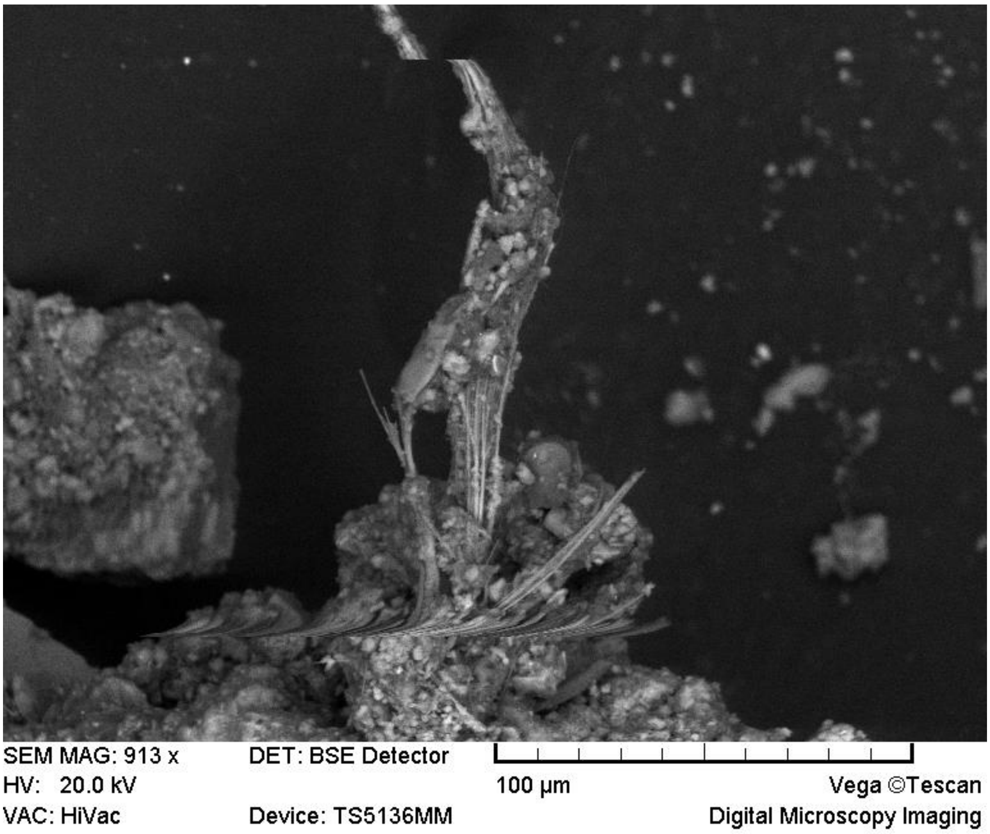

Figure 8. SEM of selected sample that contain asbestos

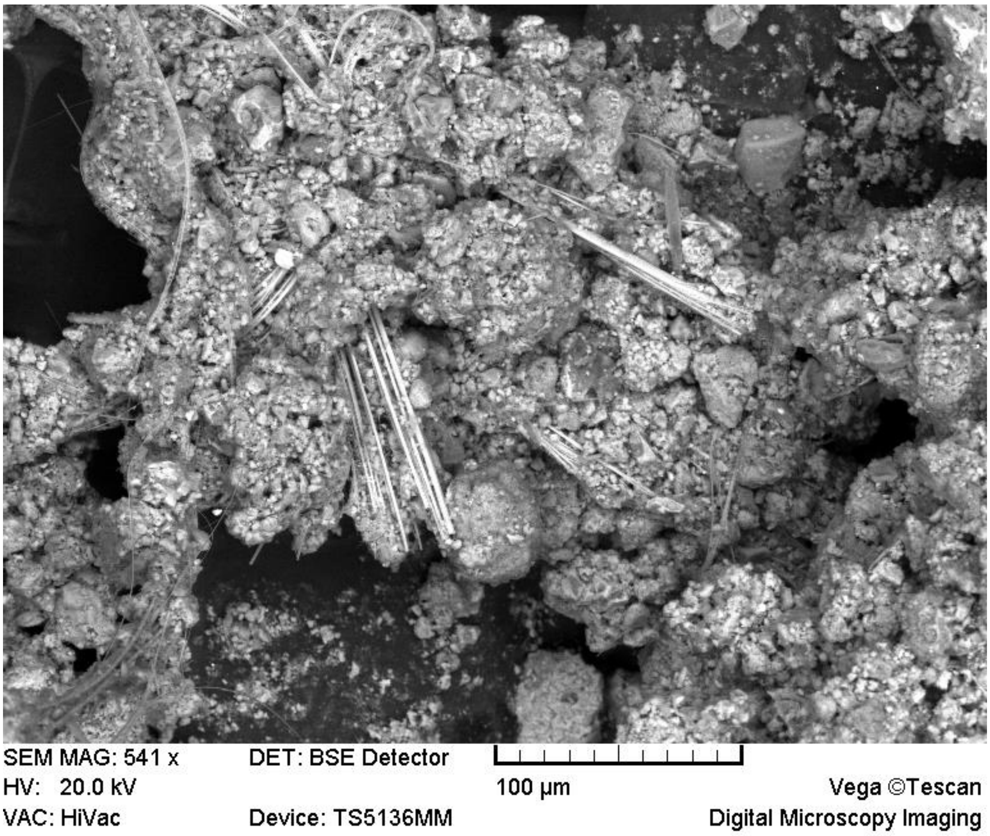

Figure 9. SEM of selected sample that contain asbestos

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АНАЛИЗА НА АЗБЕСТ ВО ГРАДЕЖНИ МАТЕРИЈАЛИ – СЛУЧАЈ ВО ГРАД СКОПЈЕ

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Азбестот е материјал кој е меѓу најтемелно проучуваните од сите материјали опасни за здравјето во минатиот век. Во нашата земја, оваа тема, за жал, не се дискутира многу иако овој проблем во светот се смета за еден од најголемите предизвици со кои треба да се справиме. Кампањата на градските власти на Скопје насочена кон откривање и елиминирање на материјалите што содржат азбест од разни предмети во градот е досега најголемата ваква иницијатива во земјата. Во неа беа опфатени методологија и резултати од анализи на примероци од градежни материјали собрани од разни објекти во градот. По систематска анализа на 50 собрани примероци од 21 јавен објект од регионот на град Скопје, со употреба на оптичка микроскопија, инфрацрвена спектроскопија, дифракција на X-зраци и СЕМ, може да се заклучи дека присуство на азбест има во 33 анализирани примероци. Важно е да се истакне дека азбестот не е идентификуван само во примероците од два објекта. Овие податоци сугерираат дека азбестот е присутен во градежните материјали на практично сите проверени објекти, што дополнително подразбира негово присуство и во многу други објекти во градот и низ земјата. Ситуацијата особено загрижува поради потврденото присуство на азбест во примероци земени од детските градинки, што е поврзано со изложеност на азбест кај младата популација.

Клучни зборови: азбест; серпетински азбест; амфиболски азбест; градежни материјали; Скопје

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Original scientific paper

NANOMATERIALS IN VOLTAMMETRIC BIOSENSORS-RECENT ACHIEVEMENTS

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While we inhabit a macro-world, it is evident that our future will depend greatly on the tiniest of things. This is due to the fact that a majority of 21st-century sciences will be centered around materials with nanometer dimensions. Currently, we observe the significance of nanomaterials in facilitating the targeted delivery of active substances within the body. In the past two years, this has played a crucial role in combating the COVID-19 pandemic, with the assistance of vaccines containing graphene oxide nanoparticles that serve as carriers and enhancers for vaccine compounds. Furthermore, challenges related to drug delivery, such as poor water solubility and limited bioavailability, have already been overcome through the utilization of metal-based and carbon-based nanomaterials. Nanomedicine is poised to revolutionize the landscape of therapeutics and diagnostics. This brief review focuses on notable achievements in designing specific voltammetric biosensors using metallic nanoparticles and graphene-based nanomaterials. Metallic nanoparticles, particularly those based on silver and gold, along with graphene derivatives such as nanotubes, quantum dots, nanodiamonds, and fullerenes, exhibit remarkable physical and chemical properties. These include improved thermal stability, enhanced conductivity, and the ability to modify their surface area with various organic substrates. Notably, voltammetric sensors based on graphene nanostructures demonstrate high biocompatibility and superior selectivity in detecting important biological systems through voltammetry. The aim of this concise review is to highlight recent electrochemical advancements in nanosystems and present significant achievements of metallic nanoparticles and graphene-based nanomaterials as voltammetric biosensors.

Key words: metallic nanoparticles; graphene; carbon nanotubes; electrochemical biosensors, voltammetry

INTRODUCTION

Voltammetry is arguably the most valuable electrochemical technique for gaining insights into various aspects of electrode transformations in physiological and chemical systems, ranging from small ions to large lipophilic proteins and enzymes [1–3]. As voltammetry revolves around measuring the energy of electrons exchanged between specific analytes and an electronic conductor (the working electrode), it becomes evident that the nature of the working electrode plays a crucial role in this electrochemical technique. A limited selection of working electrodes, primarily composed of noble metals, mercury, or carbon materials, have been reported as suitable materials for the majority of voltammetric studies [1, 4–6]. The effective utilization of voltammetry for probing specific ions, drugs, or physiologically active systems at micromolar or lower concentrations often necessitates the modification of working electrode surfaces with materials possessing superior conductive and chemical properties compared to unmodified electrodes [7]. Over the past 20 years, extensive exploration has been carried out on various nanomaterials to enhance the surfaces of electronic conductors used in voltammetric experiments [8]. Among these, nanoparticles based on metals (such as Au, Ag, Pt), metal oxides (primarily Fe_2O_3, Al_2O_3, CuO, CoO, MoO_3, Bi_2O_3), and particularly those derived from different carbon materials (multiwalled and single-walled carbon nanotubes) have been widely employed in voltammetric systems. Their usage has significantly contributed to the successful application of voltammetry in the design of numerous voltammetric sensors for detecting important chemical and physiological compounds [1, 5]. This brief overview highlights some of the key achievements of nanomaterials in the development of voltammetric sensors in recent years.

RESULTS AND DISCUSSION

The concept of "nanotechnology," initially developed by Richard Feynman [9] (Nobel Prize laureate in 1965), has brought about revolutionary advancements in the fields of nanomedicine, pharmacy, chemistry, physics, high-tech industry, food industry, and numerous other related fields [8, 10–12]. Nowadays, nanomaterials have become ubiquitous in every aspect of our lives. Over the past 20 years, nanotechnology has made significant impacts in areas such as drug delivery, cancer therapy, drug synthesis, cellular phones and computer technology, development of new materials, high-precision electronics, and environmental analysis. Although nanoparticles (NPs) encompass a wide range of materials, they are commonly defined as "three-dimensional materials with at least one dimension in their structure measuring less than 100 nm" [11]. Numerous excellent books have been published in the last 15 years, covering important topics related to the synthesis, properties, functions, characterization, and applications of various NPs. Interested readers are advised to refer to some of these publications [13–20]. In general, the remarkable characteristics of nearly all nanoparticles (NPs) that render them highly suitable for implementation in electrochemical systems are attributed to their exceptional catalytic properties, larger surface area-to-volume ratio achieved by reducing their size, improved electronic conductivity, enhanced durability, and compatibility with various substrates. Additionally, many NPs, particularly those based on graphene, serve as favorable platforms for attaching functional groups, which is an important feature manifested in their reactivity towards specific substrates. While numerous recent excellent reviews cover different properties, synthesis protocols, characterization, and application of nanoparticles in electrochemical experiments [21–26], this overview focuses on highlighting only a selection of the most significant properties and achievements of certain types of nanoparticles utilized in voltammetric biosensing. Figure 1 illustrates a scheme showcasing some of the key NPs employed in electrochemistry, while Figure 2 depicts a modified working electrode with gold nanoparticles, suitable for voltammetric experiments.

The initial mention of metal nanoparticles in electrochemistry dates back more than 30 years [11]. Metal nanoparticles are highly attractive for modifying various electrode surfaces due to their easy and cost-effective preparation, as well as their notable electrocatalytic properties [11, 14, 26]. When attached to the working electrode, these nanoparticles significantly increase its active surface area due to their small size. Furthermore, the use of metallic nanoparticles commonly enhances the material's electrical conductivity, leading to an increased rate of electron exchange between the studied analytes and the modified electrodes [4–8, 14]. Among the metallic nanoparticles investigated in voltametric studies, silver and gold nanoparticles are the most well-known [14, 21]. This is primarily attributed to their relatively inexpensive and straightforward synthesis protocol, often involving chemical reduction from salt solutions using mild reductive substances such as citric acid [11, 14].

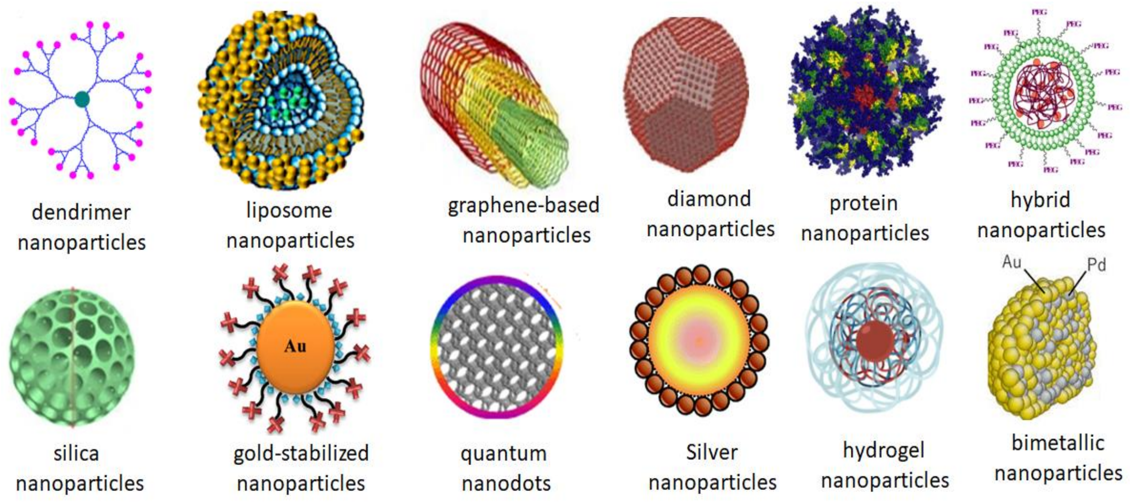

Figure 1. Schemes of some nanoforms mostly used in electrochemical experiments

Figure 2. Schematic representation of a working electrode, with active electrode surface modified with gold nanoparticles

Another advantage of utilizing silver and gold nanoparticles is their potential for stabilization and modification with various ligands and functional groups on their surfaces [26]. In the case of modifying working electrodes with silver nanoparticles, electrodeposition under controlled potential is also feasible and easily achievable, as reported in [14–17]. When silver nanoparticles are combined with graphene oxide, the reduction of silver nitrate from a water solution is commonly accomplished using hydrogen iodide as a reducing agent [27]. A chemical reduction protocol allows the production of silver nanoparticles with sizes ranging between 10 nm and 25 nm [28]. On the other hand, electrodeposition of silver nanoparticles typically results in larger particles with diameters ranging between 30 and 50 nm [27]. Unfortunately, aggregation of deposited silver nanoparticles into larger clusters often occurs and is difficult to avoid when using the electrodeposition method. The toxicity of silver nanoparticles is recognized as a significant drawback when applying these materials in experiments involving physiological systems. Gold nanoparticles are among the most extensively studied metallic nanomaterials for electrochemical detection of different analytes [29, 30]. The most common method for obtaining gold nanoparticles is through chemical reduction of chloroauric acid in water solutions using mild reducing agents such as sodium citrate, ascorbic acid, and sodium borohydride [11, 14]. An advantageous characteristic of gold nanoparticles is their ability to form covalent bonds with substrates containing thiol (-S-H) groups in their structure [14]. Numerous modification protocols involving "mercapto substrates" [31] have paved the way for the widespread use of thiol-modified gold nanoparticles in voltammetric sensing, particularly for the quantification of important biomolecules like DNA [32]. Other metallic and bimetallic nanoparticles derived from bismuth, platinum, copper, nickel, cobalt, titanium, palladium, mercury, and various other metals have also been extensively employed in various voltammetric studies [13–15]. In addition to metallic nanoparticles, metal-oxides and metal-sulfides nanoparticles have been extensively explored for voltammetric purposes over the last 10 years. Detailed information regarding their synthesis protocols, properties, functions, and applications can be found in comprehensive reports such as [14, 33–36]. Table 1 summarizes some of the most significant applications of metallic, metal-oxides, and metal-sulfides nanoparticles in voltammetric sensing of chemical and physiological systems, as published in recent years.

A significant turning point in experimental voltammetry occurred approximately 20 years ago, coinciding with the isolation of freestanding graphene [51]. The integration of this two-dimensional carbon material in electrochemical experiments marked the beginning of a new era in the development of voltammetric sensors for probing important chemical and physiological systems [52]. Due to its ability to be easily wrapped and rolled, graphene became a fundamental building block in the design of various carbon-based nanomaterials, including carbon single-walled and multi-walled nanotubes, fullerene, nanoplatelets, and other nanoforms [53]. Over the past 15 years, graphene has consistently demonstrated superior performance compared to existing electrode materials when used as a modifier for electrochemical sensors. Analysis of voltammetric sensors published since 2012 indicates that more than 30% of the works report the exploration of graphene-based nanomaterials (primarily single-walled and multi-walled carbon nanotubes) for the modification of electrode surfaces [51].

The extensive exploration of 3D graphene-based nanoforms in voltammetric biosensors can be attributed to their remarkable features, including their exceptional electrical conductivity, high chemical stability, and their ability to serve as platforms for attaching various substrates with specific functional groups onto their surfaces [54].

Table 1. Data about voltametric detection of different substrates with metal-based nanoparticles

Type of nanoparticles	**Voltammetric Technique/ Working Electrode**	**Analyte**	**Detection limit of the analyte**	**Reference**
Au-nanoparticles stabilized with dithiothreitol and dodecanethiol	DPV/Au electrode	epinephrine	0.06 μmol/L	[37]
various Au-nanoparticles	DPV and SWV/GCE/Au electrode	various polyphenols	in sub-micromolar range	[38]
Au-nanoparticles	CV/GCE	glutathione	0.7 pmol/L	[38]
Au-nanoparticles stabilized with various substrates	DPV, SWV and CV /GCE/Au electrode	DNA	sub-micromolar concentrations	[32]
various metallic nanoparticles	DPV, SWV and CV/GCE/Au electrode	hazardous polyphenols in water	sub-micromolar concentrations	[40]
Ag-nanoparticles/multi-walled nanotubes	DPV/GC electrode	glucose	0.01 mmol/L	[41]
Pt-Ag nanoflowers	DPV/GC electrode	hydrogen peroxide, glucose	0.02 mmol/L	[42]
various metallic and bimetallic nanoparticles	DPV, SWV and CV /GCE/Au/Pt electrode	Sb^{2+}, As^{5+}, Cd^{2+}, Cr^{6+}, Cu^{2+}, Pb^{2+}, Hg^{2+}, Ni^{2+}	mainly in sub-micromolar concentrations	[33]
Au-nanoparticles	DPV/GCE	glucose and hydrogen peroxide	for glucose 0.39 μmol/L for hydrogen peroxide 0.136 μmol/L	[43]
CuO nanowires	CV/copper foam	glucose	lower mmol/L range	[44]
Au-Pd/MoS_2	CV/GCE	glucose and hydrogen peroxide	for glucose 0.16 μmol/L for hydrogen peroxide 0.40 mmol/L	[45]
Pd-nanoparticles on Co-wrapped carbon nanotubes	CV/GCE	hydrazine	0.07 μmol/L	[46]
Au-nanoparticles stabilized with mercaptoundecanoic acid	CV/SWV/GCE	vitamin E	0.25 μmol/L	[47]
Au-nanoparticles stabilized with mercaptoundecanoic acid	CV and SWV/GCE	DNA	7. 5 μmol/L	[47]
Ag-nanoparticles at water-nitrobenzene interface	SWV and CV/GCE and PIGE	inorganic anions transferred across liquid-liquid interface	0.1 mmol/L	[48]
Ag/MoS_2 hybrid nanoparticles	CV/ITO	dopamine	lower mmol/L concentration range	[49]
Fe_3O_4 nanoparticles-polyvinyl chloride	CV/Sn electrode	glucose	8 μmol/L	[50]

* CV-cyclic voltammetry: SWV-square-wave voltammetry: DPV-differential pulse voltammetry:
** GCE-glassy carbon electrode: PIGE-paraffin impregnated graphite electrode: ITO-indium tin oxide electrode

To harness the extraordinary features of carbon nanotubes (CNTs) for the development of voltammetric sensors, they typically undergo initial functionalization with biocompatible molecules [55]. This functionalization step yields a biomodified nanointerface that serves as a crucial platform for designing specific voltammetric biosensors. Various organic or inorganic substrates are commonly employed for the functionalization of CNTs, resulting in improved properties such as enhanced chemical compatibility and increased water solubility [53–55]. "Covalent functionalization" primarily involves small molecules that induce changes in the π-electronic framework of graphene [56]. On the other hand, "noncovalent functionalization" of these nanomaterials involves interactions between graphene's structure and aptamers, enzymes, polymers, and other organic systems (as illustrated in Figure 3), typically achieved through van der Waals interactions [56].

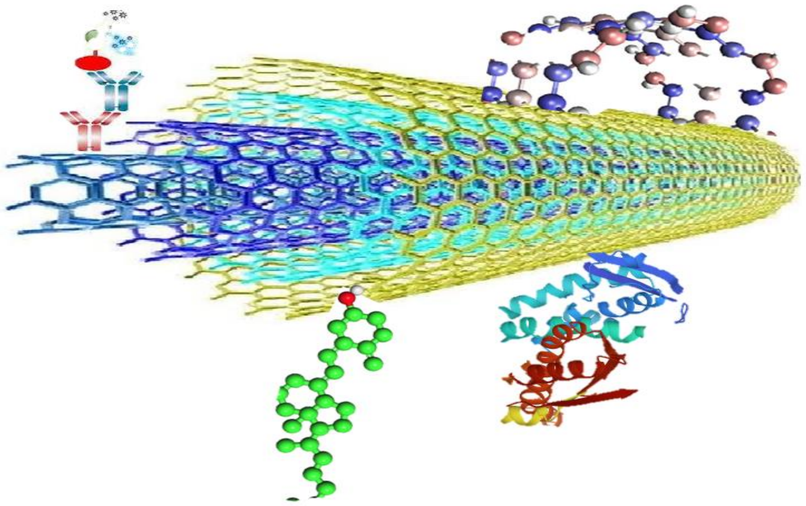

Figure 3. Scheme of a multiwalled carbon nanotubes that are functionalized with various organic molecules. The functionalization of carbon nanotubes with organic molecules is seen as a key step in achieving better selectivity of carbon nanomaterials.

Carbon nanoparticles, including graphene-made nanoparticles, have a propensity to induce changes in the electrical and chemical properties of non-modified electrodes. This often leads to significantly improved overall electrochemical performance of nano-modified electrodes for the detection of specific target molecules [56]. Over the past two decades, tens of thousands of papers have been published on electrochemical biosensors utilizing graphene-based nanoparticles in various forms, such as amperometric enzyme electrodes, voltammetric immunosensors, and nucleic acid voltammetric devices. In recent years, several comprehensive reviews have focused on graphene-based nanomaterials in the design of voltammetric biosensors suitable for quantifying diverse analytes, including DNA, dopamine, glucose, various hem-containing redox proteins, cytochromes, hormones, ascorbic acid, hydrogen peroxide, bilirubin, various pharmaceuticals, and other physiological systems. Interested readers are encouraged to refer to these works [52–61].

Enzyme-based electrochemical biosensors find extensive use in pharmacy, nanomedicine, food safety, and studies related to monitoring different substances relevant to environmental protection [53–56, 58].

Numerous voltammetric biosensors utilizing CNT-modified electrodes provide valuable insights into the activity of various redox enzymes [see reviews 58, 62, 63, and references therein]. Clinical biochemistry, being a major application area for biosensors, has witnessed a plethora of studies dedicated to the development of voltammetric biodevices for monitoring glucose, hemoglobin, urea, and other relevant physiological systems in whole blood [see reviews 58, 62–64, and references therein]. Additionally, over the past 10 years, there has been intensive exploration of graphene-based nanomaterials in designing reliable voltammetric biomarkers for medical diagnostics [65–67]. Table 2 provides information on some of the noteworthy recent achievements in the design of voltammetric biosensors using various graphene-based nanomaterials.

Table 2. Data about voltammetric detection of different substrates with graphene-based nanoparticles

Type of graphene-based nanoparticles	Voltammetric Technique/ Working Electrode	Analyte	Detection limit of the analyte	Reference
Multiwalled carbon nanotubes functionalized with cysteamine	CV/GCE	hemoglobin	0.03 μmol/L	[68]
Multiwalled carbon nanotubes immobilized on gelatin + glucose oxidase	CV/GCE	glucose	0.5 mmol/L	[69]
Multiwalled carbon nanotubes on poly(vinyl alcohol)+ alcohol dehydrogenase	CV/GCE	ethanol	0.16 μmol/L	[70]
multiwalled carbon nanotubes	CV/GCE	Cholesterol	0.5 μmol/L	[71]
multiwalled carbon nanotubes-metal oxide nanoparticles-7, 7, 8, 8-tetracyanoquinodimethane	CV/GCE	xanthine	0.2 μmol/L	[72]
composite of dendrimer-encapsulated Pt nanoparticles and carbon nanotubes	CV/GCE	Ascorbic acid, hydrogen peroxide	10 μmol/L 50 μmol/L	[73]
multi-copper enzyme co-adsorbed at carbon nanotube	CV/GCE	Hydrogen peroxide	lower micromolar range	[74]
dehydrogenase modified quantum dots-carbon (ZnS-CdS) nanotubes	CV/GCE	glucose	10 μmol/L	[75]
Hybrid of silver multiwalled carbon nanotubes/manganese dioxide.	CV/GCE	carcino-embryonic antigen	10 μg/L	[76]
Multiwalled carbon nanotubes	CV/GCE	paracetamol ibuprofen	1 μmol/L 1 μmol/L	[77]
carbon black nanoparticles	CV/GCE	hemoglobin	lower micromolar range	[78]
carbon black nanoparticles	CV/GCE	various hem-containing proteins	below 1 μmol/L	[79]
epoxy polymer and acetylene black nanoparticles	CV/GCE	Cytochrome P450	below 1 μmol/L	[80]

OUTLOOKS FOR THE FUTURE

It is increasingly evident that the remarkable properties of metallic nanoparticles and the exceptional chemical performances of graphene-based nanomaterials will play a pivotal role in shaping scientific advancements in medicine, chemistry, pharmacy, physics, new materials design, and environmental analysis throughout the 21st century [58]. During the last three years of the Covid-19 pandemic, it has become evident that the efficient functioning of vaccines relies on the support of graphene-based nanomaterials [81]. Furthermore, the delivery of many important drugs in medicine is closely linked to the unique properties of carbon nanotubes and fullerenes [82–86]. The remarkable electrochemical properties of graphene-based 3D nanomaterials have ushered in a new era of their widespread use in designing voltammetric biosensors for the detection of crucial biomolecules [58, 87]. As over 50 % of novel works on voltammetric biosensors incorporate the use of nanomaterials, we anticipate a rapid breakthrough in the more extensive application of voltammetry in commercially designed devices [88].

To enhance the sensitivity and selectivity of nanomaterials, the development of novel nanomaterials with functionalized surfaces, incorporating multiple substrates within a single nano-platform, is anticipated. This advancement will enable the simultaneous and selective determination of different biomolecules in real-time [89]. From this perspective, it is highly likely that the remarkable chemistry of nanomaterials will soon lead to the emergence of novel fields in electrochemical research that will impact various aspects of our daily lives, particularly by improving biochemical, pharmaceutical, and medical research in numerous ways [81, 83, 90–92].

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ПРИМЕНА НА НАНОМАТЕРИЈАЛИ ВО ВОЛТАМЕТРИСКИТЕ БИОСЕНЗОРИ – ПРЕГЛЕД НА НЕОДАМНЕШНИ ПОСТИГНУВАЊА

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Иако опстојуваме во макросвет, сепак, иднината на сите нас ќе зависи од работи што имаат мали димензии. Тоа е поради фактот што огромен дел од науката во 21 век е насочена кон проучување материјали со нанометарски димензии. Така, на пример, огромен дел од науката за наноматеријалите во овој период е насочена кон нивната способност за зголемување на ефикасноста на транспорт на активни супстанции во човечкото тело. Како што е познато, во минатите три години, овие наноматеријали имаа суштинска улога во борбата против пандемијата на ковид-19, при што голем дел од вакцините против ова заболување се базираа на наночестички што содржат графен оксид. Покрај тоа, голем број предизвици во медицината и фармацијата што се поврзани со биодостапноста на лековите и со нивната ниска растворливост во вода се надминати со употреба на различни наноматеријали базирани на метали или на јаглерод. Наномедицината, на пример, е гранка што ќе донесе револуционерни промени во дијагностичките и терапевтските третмани на пациентите. Во овој краток прегледен труд, главен фокус е ставен на некои од значајните постигнувања во дизајнирањето специфични волтаметриски биосензори што се базирани на метални наноматеријали и наноматеријали што содржат графен. Металните наночестички, посебно оние што содржат злато или сребро, во својот состав, заедно со наноматеријалите што се деривати на графен, како што се нанодијаманти, нанотуби и фулерени, покажуваат необични физички и хемиски својства. Така, на пример, овие наносистеми покажуваат зголемена термичка стабилност, подобрена топлинска и електрична спроводливост и имаат потенцијал за модификување на нивната површина со разни органски супстрати. Притоа, важно е да се нагласи дека волтаметриските сензори што се базирани на графенски наноматеријали покажуваат висок степен на биокомпатибилност и сензитивност кон важни биолошки системи чии својства се испитуваат со волтаметриски техники. Целта на овој краток прегледен труд е да ги прикаже некои од најновите електрохемиски придобивки од наноматеријалите и да ги претстави некои од значајните постигнувања на метални наночестички и наноматеријали базирани на графен во сферата на волтаметриските биосензори.

Клучни зборови: метални наночестички; графен; јаглеродни наноцевчиња; електрохемиски биосензори; волтаметрија

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Original scientific paper

APPLICATION AND IMPORTANCE OF SCANNING AND TRANSMISSION ELECTRON MICROSCOPES IN SCIENCE AND TECHNOLOGY

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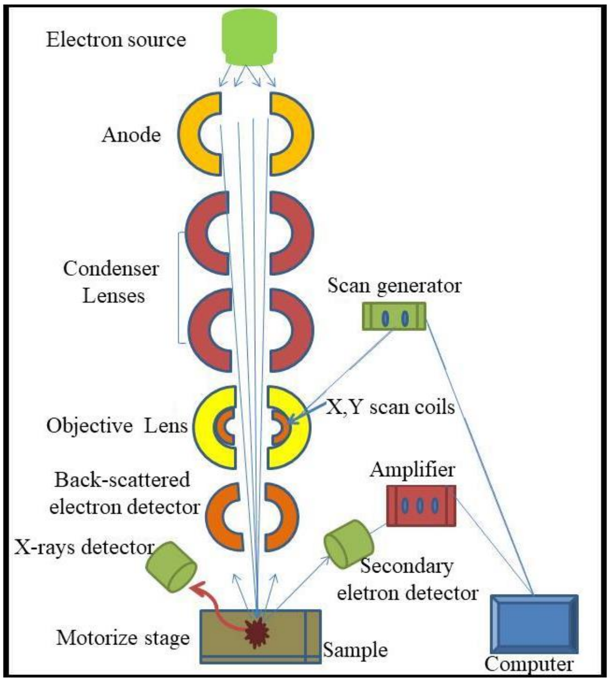
Recent developments in nanoscience and technology have increased the importance of electron microscopes, specifically the scanning electron microscope (SEM) and transmission electron microscope (TEM), in almost all fields of science and technology. Electron microscopes provide information such as morphology, composition, crystal structure, and crystal phase. They have the capability to confirm the formation of required products and also provide information on any defects that may occur in the product. SEM and TEM have been used in various disciplines including chemistry, physics, biology, pharmacy, archaeology, materials science, agriculture, soil science, environmental science, forensic sciences, civil engineering, and electronic engineering. All these features and advantages of electron microscopes contribute to their importance in academic as well as industrial research, thereby enriching the development and growth of nanoscience and nanotechnology.

Key words: SEM; TEM; nano-materials; nano-science; nanotechnology

INTRODUCTION

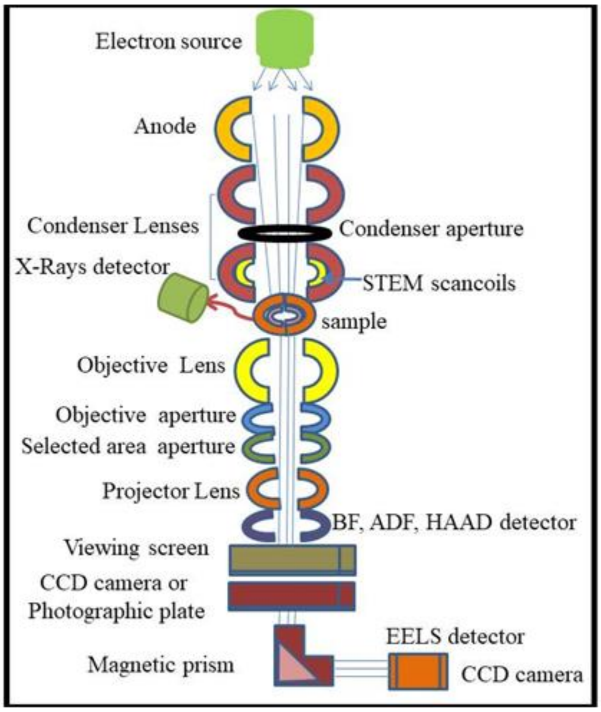
Nano-science, nanotechnology, nanostructure, and nanoparticles are now very common and frequently used terms in the literature of all fundamental sciences and technologies. Nano-science and nanotechnology pertain to the creation, study, and application of systems, materials, and devices that can control matter in the range of 1–100 nm for the betterment of the human future. The concept of nanotechnology can be referred to as the construction of miniature machines, ultra-precision processing, robots, supercomputers, molecular devices in the nanometric range, and their application in science and technology. This nanorange strongly affects their properties and behaviour [1]. Thus, in the current era, nanotechnology is considered the superior technology for all fields of study. In this regard, the investigation of materials at the nano level is highly recommended to understand their properties. However, there are serious challenges in the production of nanotechnology-based products, such as controlling size, distribution, uniformity, crystal structure, lattice type, morphology, and confirming these aspects, which require highly sophisticated characterization techniques [2, 3]. The scanning electron microscope (SEM) and transmission electron microscope (TEM) are highly sophisticated techniques in science and technology that provide morphological, elemental, and crystallographic information about a sample. They have been developed into vital tools for a better understanding of material properties and interpreting their behavior [4, 5].

The scanning electron microscope is actually based on the surface scanning of a sample with the help of a focused electron beam, while the analysis is made from the signals obtained from the interaction of electrons with the sample [4], a schematic diagram is shown in scheme 1. The electronic interaction in the scanning electron microscope helps to develop images with high magnification and large resolution, surpassing those achieved by a light microscope due to the distinct wavelengths of light and electrons. SEM also possesses the capability to analyze the chemical composition of a sample, enabling qualitative and quantitative measurements of elements [6, 7]. In the SEM, electrons are generated in an electron gun and then focused on the sample with the help of a condensing lens and beam-defining aperture to fall on the sample. Once these electrons strike the sample with their high energy, they eject secondary electrons from the sample. These secondary electrons are captured by a detector and converted into an image with the help of a computer. They provide very important information about the sample's surface morphology, crystallography, composition, and other properties. The SEM has high resolution, which may be due to the electron beam diameter and excitation volume. Similarly, electrons produced with high energy experience elastic collisions with the atomic nuclei of the sample, resulting in backscattered electrons. Backscattered electrons are extremely high-energy negatively charged particles that are usually used to produce images with greater pixel density and resolution, showing the dispersion of numerous elements contained in a specimen. Consider an exoplanet that rotates around a planet but then starts moving back into space again without being affected by the planet's gravitational effects. Similarly, whenever the specimen is exposed to high-energy electrons, a portion of these negatively charged subatomic particles will not be absorbed by the positively charged nucleus due to the influence of its centripetal force and outward pull of the atom. Instead, they will be back-reflected, resulting in "backscattered" electrons from the specimen. The amount of scattering electrons produced varies with the specific atomic mass of each element. Therefore, elements with larger atomic weight and larger nuclei, which correspond to denser materials, significantly deflect incident electrons more strongly than smaller components. As a consequence, heavy nuclei such as silver (Ag) with an atomic number (Z=47) appear brighter in SEM micrographs compared to light nuclei, such as silicon (Si) with an atomic number (Z=14), even though a large number of backscattered negatively charged particles are emitted from the surface of the specimen. These electrons are detected by a specialized detector and provide information about the local crystallographic structure of the sample. Additionally, when electrons are ejected, they also generate X-rays, which can be utilized for quantitative and qualitative analysis of the elements present in the sample. This technique is known as X-ray microanalysis, also referred to as wavelength-dispersive spectroscopy (WDS) or energy-dispersive spectroscopy (EDS). In SEM, structural defects and trace element composition can be confirmed by examining the light emitted from the sample, which is a result of the cathodoluminescent phase. This analysis is known as cathodoluminescent (CL) imaging analysis. High resolution and nanoscale investigation are the most attractive benefits of SEM. However, SEM is not recommended for samples that require high pressure or special preparation protocols due to their composition. The environment inside the column of SEM and TEM plays a crucial role in these electron microscopes. The production or monitoring of the electron beam in TEM and SEM relies on maintaining adequate vacuum. The presence of oxygen, nitrogen, or other particles can significantly reduce the operating life of the filament. This can be likened to allowing air into a tungsten filament of a light bulb, which would cause it to burn out. Although these gaseous particles in the SEM and TEM vacuum chamber can also serve as samples, they diffuse the intensity of the electron beam when struck by the negatively charged electrons. Recently, environmental SEM (ESEM) has been developed to address this issue for sensitive samples, such as biological samples. ESEM has the capability to maintain the sample at moderate pressure while keeping the electrons at high pressure, allowing for easier study of the sample. This requires special protocols in SEM [6, 8].



Scheme 1. Schematic diagram of the core components of an SEM

The transmission electron microscope (TEM) is designed similarly to a light transmission microscope, with the only difference being that light is replaced by electrons [9]. In TEM, electrons are generated in an electron gun and then focused on the sample using condenser lenses and an aperture. The electron beam strikes the sample and transmits through its thin surface, creating an image on the objective lens. This image is further magnified by passing it through the projector lens, which adjusts its strength. In the case of a crystalline sample, a diffraction pattern is formed in the lens known as the back focal plane [7]. A schematic diagram illustrating these concepts is shown in Scheme 2. Dark field (DF) and bright field (BF) imaging techniques form the basics of TEM. High-resolution TEM (HRTEM) allows for the examination of atomic structures and crystal information through the interaction of electrons with the crystal planes of the sample. Additionally, selected area electron diffraction (SAED) provides valuable insights into the crystal structure and the presence of crystalline or amorphous regions in the sample.



Scheme 2. Schematic diagram of the core components of a TEM

In order to define the sample area, scanning transmission electron microscopy (STEM) can be applied, and energy-dispersive X-ray spectroscopy (EDS) and Electron Energy Loss Spectroscopy (EELS) can provide comprehensive information for qualitative and quantitative elemental analysis of the sample, similar to SEM. Similarly, chemical analysis can be performed using energy-filtered TEM (EFTEM), where electrons with specific kinetic energies are employed for investigative study. To utilize these techniques effectively, a complete and detailed understanding of the sample is necessary in order to better comprehend its composition, texture, structure, morphology, and properties for various applications [7, 9–11]. TEM has the capability to serve as a complementary characterization technique to X-ray diffraction (XRD) and provide valuable crystal information that can support XRD data. However, TEM can be considered superior to XRD due to its additional features. In addition to crystal information, TEM can provide comprehensive information about the morphology and chemical composition of a sample, both qualitatively and quantitatively. It can provide insights into crystalline, pseudo-crystalline, and amorphous samples, similar to XRD [12–15]. Additionally, electron tomography is a technique that utilizes TEM to generate a three-dimensional image of a targeted sample by capturing images at different tilt angles around a single axis. This technique is particularly significant in paleontology as it enables the reconstruction of the internal structure of microorganism fossils [8]. The primary distinction between SEM and TEM lies in their imaging mechanisms. SEM generates images by scanning the surface of the specimen and detecting reflected, bounced-back, or knocked-off electrons. On the other hand, TEM generates images by measuring the density of transmitted electrons that pass through the specimen. As a result, TEM provides crucial information about the internal composition of the specimen, including lattice parameters, topography, and stress conditions. In contrast, SEM provides insights into the external surface and stoichiometry of the specimen.

KEY APPLICATIONS OF ELECTRON MICROSCOPES

Electron microscopes possess the capability of high resolution, allowing for the determination of crystal structures as well as the aggregation of atoms in various nanostructures such as nanowires, clusters, nanoparticles, and thin films. They are also adept at detecting and characterizing phenomena such as cracking, dislocations, and surface contamination in samples. Furthermore, electron microscopes provide valuable information about polycrystalline and ceramic materials [16].

SEM can be applied to conduct a detailed topography study of a sample, providing microstructural information such as material size, distribution, and density. It also offers valuable insights into morphology, particularly in terms of 3D morphology, including the number and shape of materials, as depicted in Figures 1a, b, 2a, b, and 3a-c.

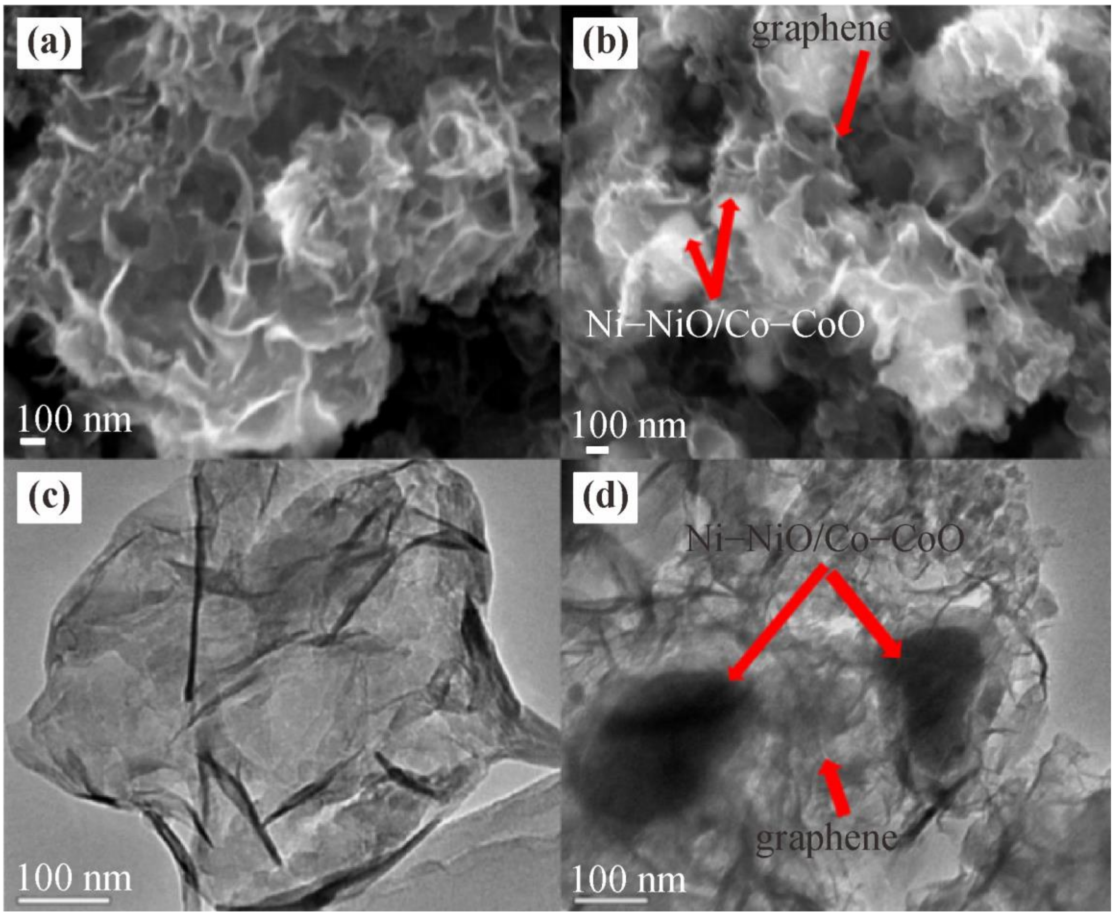


Fig. 1. (a) SEM (b) TEM and (c) HRTEM images of Ni-GCS. Reproduced with permission from Ref. [17] (Copyright 2020, Springer.)

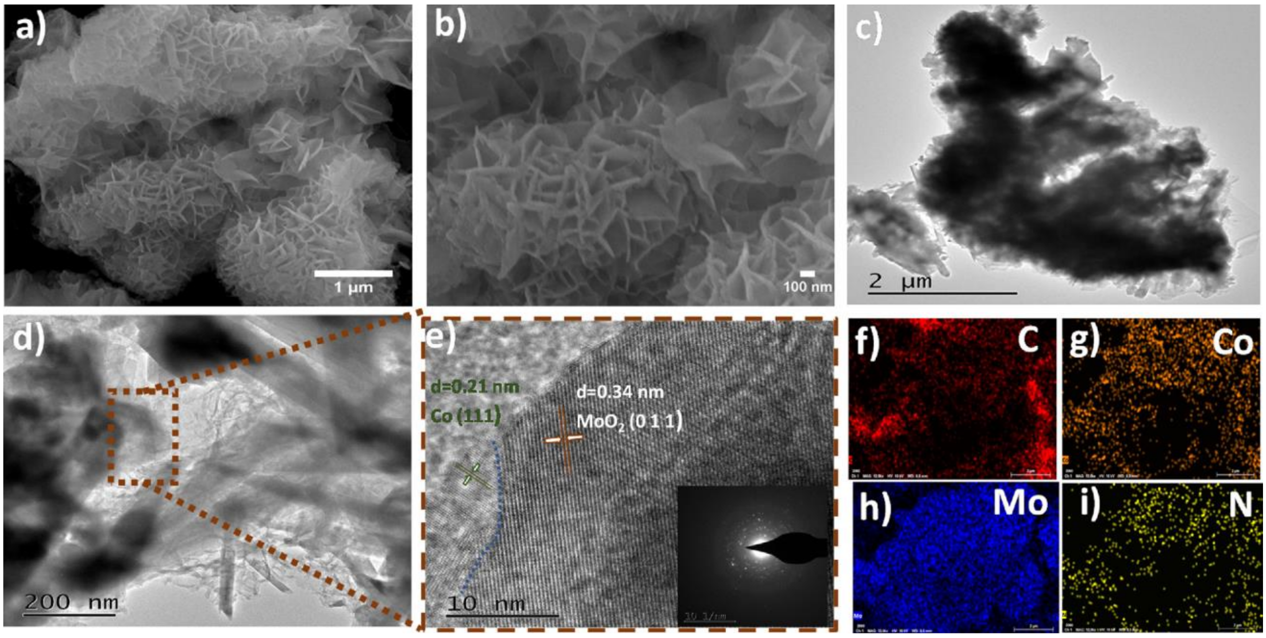


Fig. 2 The morphology and crystal structure of CMO@NC/450 sample: (a, b) SEM, TEM and HR-TEM images (inset showed corresponding SAED pattern); images; (d-e) and (fei) EDS elemental mapping (Scale bar: 2 mm). Reproduced with permission from Ref.[18] (Copyright 2020, Elsevier B.V.).

Moreover, SEM facilitates the examination of porosity within the sample, including its size, tortuosity, and distribution. It is also useful for investigating cracks, including their size, length, shape, and interconnection. Additionally, SEM, aided by energy-dispersive spectroscopy (EDS) as shown in Figures 2f-i, enables qualitative and quantitative analysis of elements present even in trace amounts in the sample. Furthermore, electron backscatter diffraction (EBSD), a specialized technique in SEM, is instrumental in determining crystal structures, orientations, morphologies, and sizes of individual grains, as well as in alloys and composites, and their crystallographic relationships with other phases. [16].

Similarly to SEM, TEM also enables chemical and structural analysis of samples with high magnification and resolution. While SEM is limited to a pixel density and resolution power of 0.5 nm, TEM has confirmed the capture of photos with ultrahigh resolution as low as 50 pm. Moreover, the magnification power of TEM surpasses that of SEM significantly. Transmission electron microscopes can enlarge specimen sizes by over 500 million times, whereas the optical zoom of SEM is restricted to 1–2 million times. Techniques with atomic resolutions such as HAADF, STEM, and HRTEM are employed in the analysis of crystal structures, interfaces of different phases, and grain boundaries, as illustrated in Figures 1c, d, 2c-e, 3d, e, and 4.

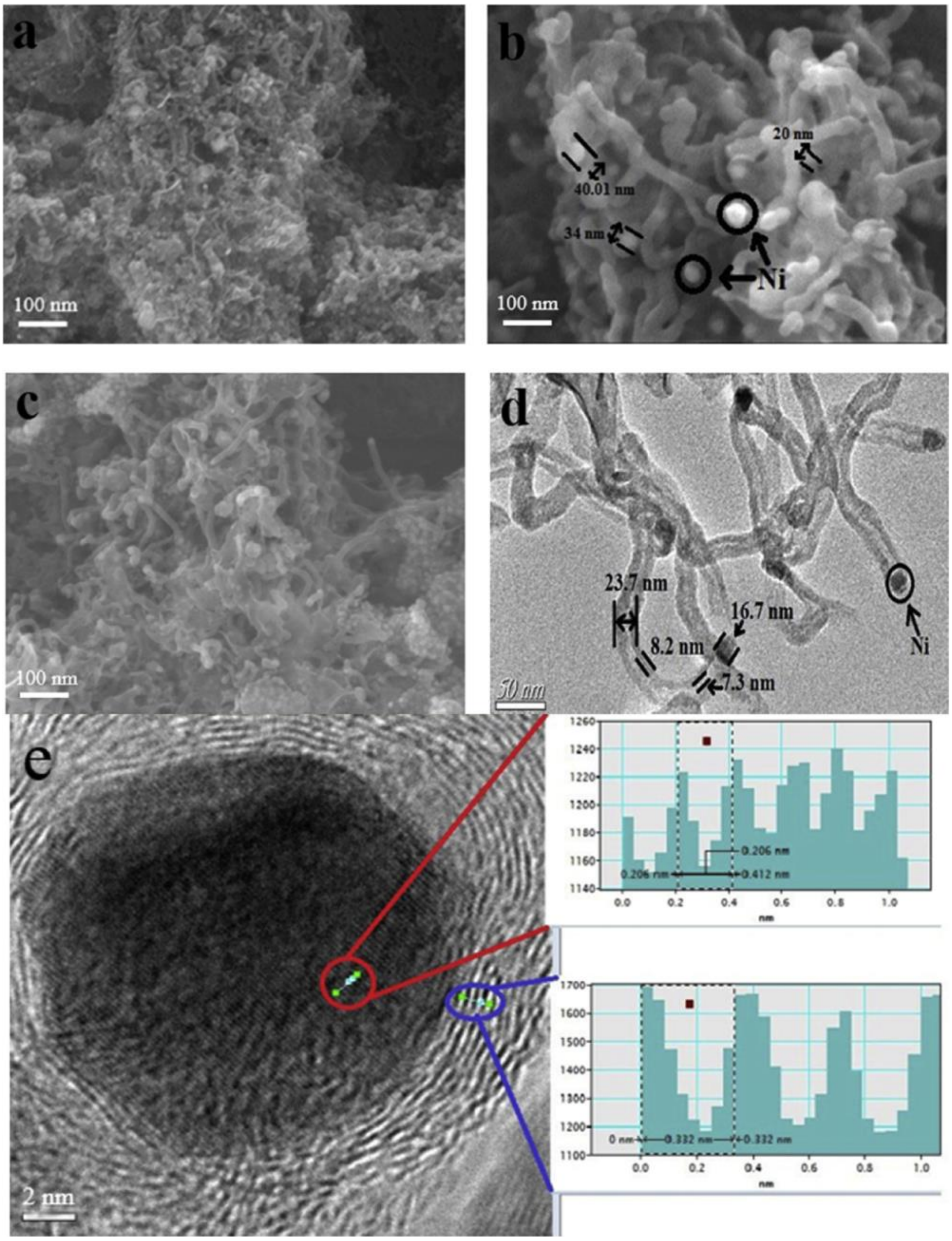


Fig. 3 (a-c) SEM image of the Ni encapsulated CNTs at 600, 650 and 700 _C
(d) TEM image of the Ni encapsulated CNTs at 650 _C (e) HRTEM image of the Ni encapsulated CNTs at 650 _C
Reproduced with permission from Ref. [19](Copyright 2020, Elsevier B.V.).

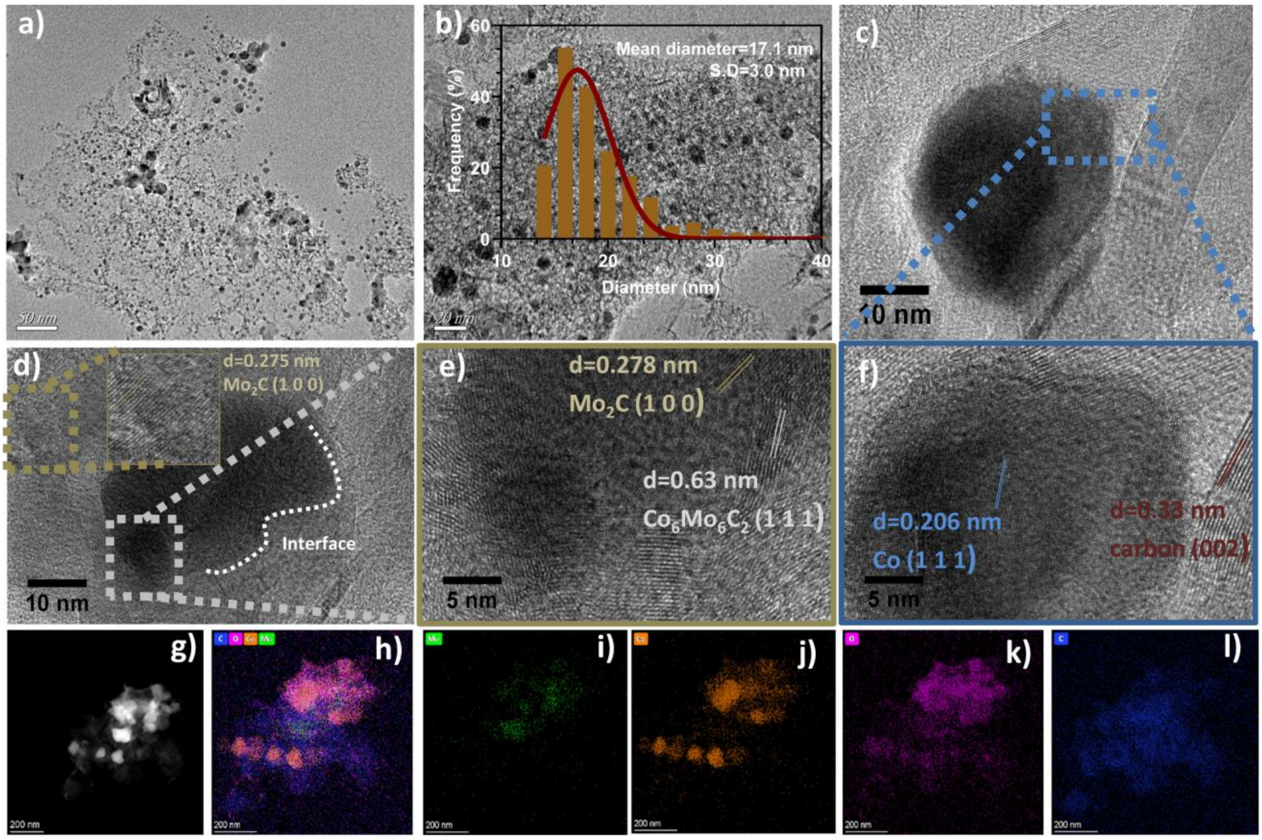


Fig. 4. (a-b) TEM image of CMC/750SA; (b) Inset: Particles size distribution of CMC/750SA; (c-f) HR-TEM image of CMC/750SA catalyst and distance between lattice fringes; (g-h) HAAF elemental mappings; (i) molybdenum; (j) cobalt; (k) oxygen; and (l) carbon (Scale bar = 200 nm). Reproduced with permission from Ref. [20] (Copyright 2020, Elsevier B.V.).

Related electron diffraction techniques can be utilized to quantitatively analyze the crystal structure and corresponding lattice parameters, as demonstrated in Figures 3e, 4e, and 5e, f. These techniques also aid in identifying imperfections such as dislocation cores and stacking faults. Additionally, EDX and EELS are employed to assess homogeneities and associated modulations, including amorphous layers in grain boundaries, precipitation, and surface impurities. Elemental mapping provides comprehensive information about the presence of all elements and their distribution, confirming the homogeneity within the sample, as shown in Figures 4g-l. TEM has the capability to determine surface morphology, including carbon sheets/graphene, nanoparticles, differentiation of carbon nanotubes and nanowires, as well as particle sizes in the sample, as depicted in Figures 1c, d and Figures 3d.

It helps in understanding the chemistry of 3D morphology and the encapsulation of materials, as demonstrated in Fig. 1d and Fig. 3e, which is particularly beneficial in minimizing leaching of active materials and in drug delivery applications. Additionally, it provides comprehensive analysis of electronic, polymeric, and semiconductive devices, as well as their related chemistry, to assess their functionality [16]. In addition, SEMs generate highly detailed 3D images of the external surface of the specimen, while TEM analysis provides 2D views of the specimen. This difference in imaging can sometimes make it challenging for researchers to correlate their findings between the two techniques, as depicted in Fig. 1 and 2. However, all characterization techniques, including SEM, TEM, and XRD, are complementary to each other and offer mutual support. SEM, TEM, and XRD provide similar information, but with different emphases. XRD primarily provides information on the composition and crystal structure of the sample, while SEM and TEM excel in providing detailed information about the topography and morphology of the sample. However, these techniques still benefit from cross-validation and support from one another to confirm sample characteristics and enhance the understanding of the sample's chemistry for its application in the field of science and technology.

Importance of electron microscopes in science and technology

In the last two decades, great advancements in nanoscience and nanotechnology have been made, which have greatly increased the importance of electron microscopes due to their effective application in nanoscience and technology.

Electron microscopes in biological and medical science

The current advancement of nanoscience, technology, and their long-term effects has also attracted the biomedical field for the synthesis of nanodrugs/nanomedicine and their application in medical sciences. In this regard, recently, 22,000 new articles focusing on nanomedicine for cancer treatment have been published, and all of these articles have utilized electron microscopes to study the medicines and their effects on cell death and necrosis. The investigative studies primarily concentrate on the morphology and structure of nanomedicine, its relationship with cytoplasmic organelles, and its impact on cell death or damage. TEM has the capability to determine the physiochemical characterization of freshly synthesized samples and assess the effects of nanomedicine on biological organisms, providing valuable information for effective therapeutic and diagnostic strategies. Thus, histochemists and biologists have always supported physicists, chemists, and pharmacologists in the synthesis of nanostructures/nanomedicine. This technique is convenient for investigating nanovector-based therapies aimed at treating affected cells that require preservation and functional repair. Following drug delivery, ultrastructural cytochemistry describes any changes in morphofunctional features, providing evidence of negative effects or restoration of healthy characteristics. All of this information requires an appropriate cell system and the support of biologists for those involved in nanoscience/technology [21]. Electron microscopes can be used to investigate histopathology and determine abnormalities in cells and tissues. Additionally, techniques such as EDX and EELS can be applied to identify significant issues in biological samples [22]. Colloidal gold particles function as specific complementary DNAs when attached to DNA [23], and DNA microchip can be used for mapping genetic information in RNA and DNA. Nanosensors can be applied to understand the effects of different stimuli on living organisms and to determine various environmental issues and their effects [24]. In addition, these techniques can be used to investigate the topography, morphology, crystallinity, and composition of newly developed drugs. They can also help in identifying any abnormalities in the new drug and quantitatively determining its ingredients, even in trace amounts. The importance and application of electron microscopes remain significant in this area of study. Sensitive (biological) samples can be treated specially and studied under electron microscopes to examine abnormalities, depositions, functions, treatments, improvements in health, side effects, and cell death, among other factors.

Electron microscopes in archeology

SEM has the capability to investigate various materials for archaeological applications, such as faience, stone, metals, glass, soil particles, pottery, bone, fingernails, hair, teeth, skin, eggshell, mollusks, wood, insects and parasites, plant remains, pollen, fibers, pigments, and more. Electron microscopes can be used to qualitatively and quantitatively determine the composition of these materials and identify decay or other structural and compositional defects. Electron microscopes can quickly determine the crystal structure and phase of the materials in a nondestructive manner. In particular, SEM does not require extensive sample preparation protocols and can provide sufficient qualitative information about the constituents of the sample. TEM can also be used to investigate the internal structure of samples, especially thin fossils or samples collected from archaeological sites, without damaging them [25].

Electron microscopes in earth sciences

Electron microscopes have the advantage of high resolution, allowing for in-depth investigation of samples. This is particularly beneficial in the field of earth science, as electron microscopes enable direct analysis of non-conductive samples without the need for pre-treatment. Electron microscopes provide detailed imaging of the sample surface, as well as analysis of its crystal structure and composition. The direct examination of samples in their natural state eliminates the requirement for pre-treatment. When combined with energy-dispersive X-ray spectroscopy (EDX) and backscattered electron imaging (BSE), electron microscopes can provide accurate information about the presence and concentration of all elements in the sample. Line scanning and mapping techniques further aid in the determination of migration patterns, mineral alteration, and growth conditions. EBSD can determine phase changes in minerals, grain orientation, and the crystal structure of the sample. Electron microscopes have been instrumental in investigating zircon and providing essential information for in-situ U-Pb dating of zircon. These microscopes have demonstrated excellent accuracy in analysis. Advanced features, such as cathodoluminescence (CL), can be employed in electron microscopes to study luminescent samples under electron irradiation. This technique provides more detailed information about trace elements, their distribution, and lattice defects. [26]. Similarly, the electron microscope has been applied to investigate the composition and quality of coal. Thanks to its capabilities, the electron microscope can be used to determine the qualitative and quantitative composition of each ingredient in coal, which serves as a basis for assessing coal quality. This qualitative and quantitative analysis is crucial in determining the production of pollutants that occur after coal combustion [8]. In addition, electron microscopes can be utilized for qualitative and quantitative analysis of various minerals to determine structural defects, composition, and assess their quality. These powerful instruments provide valuable insights into the internal structure and characteristics of minerals, enabling researchers to study their properties in detail and make informed assessments of their quality.

Electron microscopes in engineering

Nanotechnology is an emerging field of science and technology that finds applications in civil engineering. In civil engineering, nanotechnology can be utilized to reduce concrete segregation, incorporate copper nanoparticles in low heat Portland cement, develop nanosensors for monitoring early-age concrete properties, and design granules for water purification [27–29]. Nanotube-based transistors and nanomaterial-based semiconductors serve as the foundation for advanced microchips, which are fundamental to current technology [30]. Additionally, nanotubes can function as intermolecular wires in electronic devices[31]. Nano base semiconductor especially III-IV nitride such as InGaN as laser diods and LEDs are impressive technology while quantum dots and wires have numerous uses [32]. To better understand the shape, morphology, surface feature, structure, composition, and its relation with its properties is superior side of civil engineering. To get deep information about structure, morphology, composition and its relationship with application can be done with electron microscope. In addition, electron microscopes can be applied to study the microchips, transistor and diodes which are the prime part of electronic engineering in current society. Electron microscopes can determine the composition and structure of microchips, transistor, diodes, and their defect if it happens to these chips during development [33]. Basically, the study of materials for the construction of these chips, transistor and diodes can be done with electron microscopes and provide its structure, composition, crystallinty, and its relationship with conductivity for electronics application.

Electron microscopes in food/agricultural science

Electron microscopes have been applied to study the strength of eggshells and their relationship with palisade (directly proportional) in the field of food science. SEM and TEM have confirmed that frozen and concentrated egg yolk has an open surface structure compared to normal egg yolk, and any change in concentration leads to a change in its shape and organization. The freezing process may aggregate lipoproteins in low and high-density egg yolk, and this aggregation may develop a 3D structure and increase its viscosity. Electron microscopes have confirmed that tomato production can be increased with increased potassium, and potassium is independent of magnesium and calcium, showing an inverse effect. It has also been confirmed that starch gelatinization takes place inside cell units, other cell components disperse, and cells separate without rupture, and denaturation of proteins causes food swelling during the cooking process. Furthermore, it has been confirmed that urea-containing food supplements are toxic if they contain 50g of urea per 100 kg of supplement [34]. In addition, the electron microscope can also confirm the composition and concentration of synthetic or natural food, thereby assessing its structure, morphology, and effects on taste and quality. The study of the relationship between different substances and the quality and quantity of food, as well as the effects of various supplements on food production, is also a part of this scientific field. In addition, the electron microscope can also confirm the composition and concentration of synthetic or natural food, thereby assessing its structure, morphology, and effects on taste and quality.

Electron microscopes in materials/chemical/physical/natural sciences

It is well known that the science and technology of nanomaterials is an interdisciplinary field of study encompassing materials science, chemistry, physics, biology, and engineering. These disciplines are interconnected, and scientists from these backgrounds must collaborate in the fields of nanoscience, nanomaterials, and nanotechnology. This technology finds applications in various sectors such as electronics, space exploration, industry, chemicals, medicine, and healthcare, benefiting humanity. Material chemists play a significant role in the design, assembly, synthesis, and catalysis of these materials [32]. The sol-gel and aerogel methods have been developed to create materials with high surface areas and exceptional absorbent properties, making them effective in the removal of toxic substances [35, 36]. Nanocomposites have resulted in the production of materials with ultrahigh toughness, strength, and ductility, which find applications in various fields such as cement, ceramics, plastics, textiles, and novel magnets. Nanomaterials, characterized by their nanosize and unique morphology, exhibit exceptional catalytic properties and are utilized in capacitors, metal-ion batteries, and other industrial applications [37]. Nanoporous materials, polymers, and nanotubes exhibit excellent applications in catalysis, storage, purification, and lubricant applications [30, 32, 38]. In order to gain a better understanding of nanomaterials, including their surface topography, morphology, and size, electron microscopes such as SEM and TEM are crucial. These electron microscopes are necessary for investigating the crystalline and amorphous structures of materials and understanding their applications in catalysis, adsorption, capacitance, and batteries. Electron microscopes also enable quantitative and qualitative analysis of materials, allowing researchers to determine their effects on various applications. They can easily investigate the agglomeration of active materials, which can impact their performance. Additionally, electron microscopes are capable of studying hetero-junctions, the interaction between metals and supporting materials, and their effects on applications. Furthermore, electron microscopes can provide valuable insights into the crystal structure, crystal phase, and atomic arrangement within crystals.

Electron microscopes play a crucial role in forensic science by allowing for in-depth investigation of samples to gather evidence. They have the capability to analyze even trace amounts of samples and provide valuable information about compounds, compositions, and concentrations. This makes electron microscopes an indispensable tool in forensic investigations. The significance of scanning electron microscopy (SEM) and transmission electron microscopy (TEM) extends beyond forensic science and extends to all fields of science and technology. Their importance cannot be denied in today's society, as they enable researchers and scientists to gain a deeper understanding of materials, structures, and phenomena at the nanoscale. Electron microscopes have revolutionized scientific research and continue to play a vital role in advancing various disciplines.

CONCLUSION

The emerging field of nanomaterials has opened new avenues in science and technology, leading to significant advancements in recent years. One crucial aspect of this field is the confirmation of required materials, and in this regard, scanning electron microscopy (SEM) and transmission electron microscopy (TEM) have gained extensive attention. These techniques provide sophisticated information by enabling investigations at the subnanoscale range and detecting trace elements in the sample. Moreover, they offer advanced insights into morphology, composition, crystal structure, and defects of the desired products. The applications of SEM and TEM span across various disciplines, including chemistry, physics, biology, pharmacy, archaeology, materials science, agriculture, soil science, environmental science, forensic science, civil engineering, and electronic engineering. Importantly, the data obtained from electron microscopy can be compared with results from other techniques for confirmation, as these techniques are complementary. The numerous features and advantages of electron microscopy highlight its significance in both academic and industrial research, contributing to the development and growth of nanoscience and nanotechnology.

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ПРИМЕНА И ВАЖНОСТ НА СКЕНИРАЧКАТА И ТРАНСМИСИОНА ЕЛЕКТРОНСКА МИКРОСКОПИЈА ВО НАУКАТА И ТЕХНОЛОГИЈАТА

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Најновиот развој на т.н. нанонауки и технологии ја истакнаа важноста на електронската микроскопија, особено на скенирачката и трансмисиона електронска микроскопија во многу области на науката и технологијата. Електронската микроскопија нуди информации за морфологијата на материјалите, нивниот состав, кристалната структура и постоењето различни кристални фази. Со оваа техника може да се докаже дека се синтетизирани нови материјали со посакувани особини, но и да се откријат дефекти во нивниот состав и структура. Скенирачките микроскопии се користат во многу научни полиња, како што се хемија, физика, биологија, фармација, археологија, наука за материјали, земјоделство, наука за почвата, наука за животната средина, форензичка наука, градежништво, електронска индустрија итн. Многуте предности на овие техники придонесуваат за нивната важност и неопходност во светот на академските истражувања, а и во индустријата, што сè заедно овозможува особен развој на нанонауката и нанотехнологијата.

Клучни зборови: скенирачка електронска микроскопија; трансмисиона електронска микроскопија; наноматеријали; нанонаука; нанотехнологија

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ADVANCED PROCESSING OF ELECTROCHEMICAL DATA IN SQUARE-WAVE VOLTAMMETRY

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The processing of data in voltammetry is crucial for extracting mechanistic and kinetic information about the electrode reaction under study. One of the most advanced pulse-voltammetric techniques, square-wave voltammetry (SWV), allows for the investigation of electrode mechanisms, measurement of fast electron transfer reaction rates, and quantitative detection of redox species at sub-micromolar concentration levels. To ensure reliable comparison and fitting of experimental and theoretical data, several ideas for appropriate data processing have been proposed, considering the differences between experimentally accessible current and the dimensionless current function used in the theory and mathematical modeling of electrode processes in SWV. Significantly, in the context of advanced and recently introduced double-sampled square-wave voltammetry, a novel methodology for data analysis has been introduced. This methodology transforms the conventional current-potential function (i.e., voltammogram) into a current-current function, analogous to complex numbers. By employing this methodology, the accessible kinetic interval is extended, allowing for the measurement of very fast, virtually reversible electrode reactions characterized by a standard rate constant as high as 0.1 cm s^{-1}.

Key words: square-wave voltammetry; simulations; processing of voltammetric data; electrode kinetics; double-sampled square-wave voltammetry

INTRODUCTION

Voltammetry is a fundamental experimental methodology in electrochemistry and related fields, designed to study the mechanism, kinetics, and thermodynamics of electron transfer (redox) reactions. It offers powerful analytical methods for both quantitative and qualitative determinations [1]. Voltammetry has a rich historical background, dating back over a century, and has evolved into its current state of development, primarily represented by pulse voltammetric techniques [2]. Pulse voltammetry is specifically designed to enhance sensitivity for the quantitative detection of redox compounds, even at trace levels. Among the multitude of pulse techniques, square-wave voltammetry (SWV) stands out as one of the most advanced methods. It combines the advantages associated with cyclic voltammetry, such as mechanistic studies, with excellent analytical performance [3] (Fig. 1). As a result, SWV has become the cornerstone of highly sensitive methods in electroanalytical chemistry [4]. The theoretical foundation of this technique has been developing since the 1960s [5, 6]. However, significant methodological advancements in both theory and experimental applications have been witnessed in recent years [7–9]. Building upon the methodological improvements proposed by Krulic et al. [10], square-wave voltammetry (SWV) has recently undergone transformations to function as a pulse-form of chronoamperometric technique. This modification aims to simplify the technique and enable advanced applications in conjunction with amperometric sensors [11]. Furthermore, a hybrid voltammetric technique has been developed by combining SWV with differential pulse voltammetry [12]. This combination enhances the effectiveness in discriminating residual current while retaining the capability to resolve electrode mechanisms and kinetics. Additionally, to enable comprehensive studies of electrode reactions within a single experiment, multisampling [13] and double-sampling [14] current protocols have been proposed. These protocols facilitate the investigation of reaction mechanisms and kinetics, as well as the advanced analysis of sluggish electron transfer processes [14].

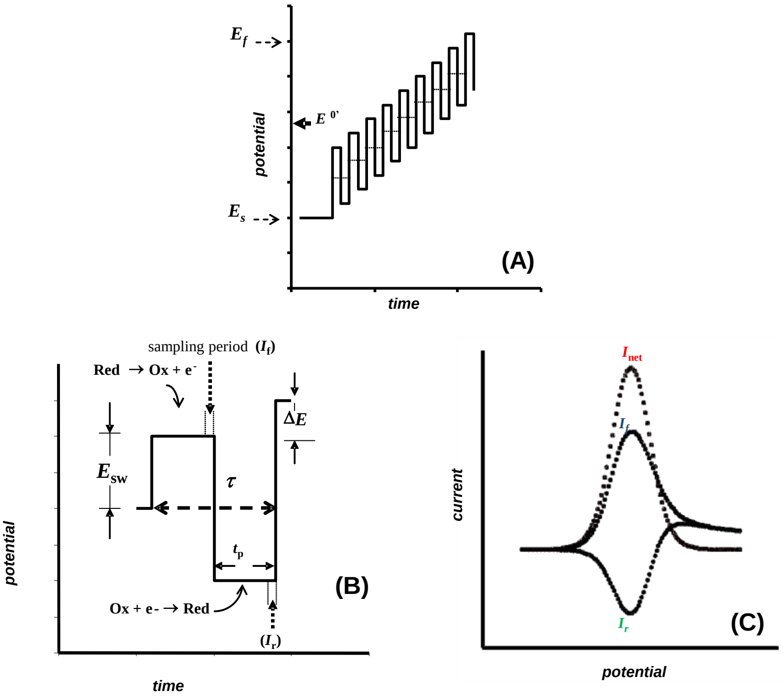


Fig. 1. (A) The potential waveform in square-wave voltammetry (SWV) can be visualized as a combination of a staircase potential and alternating potential pulses, overlaid on each step of the staircase waveform. **(B)** In SWV, a single potential cycle with a duration of τ comprises two opposing pulses of equal magnitude (E_{sw}, defined as the SW amplitude), each with a duration of t_p, such that $\tau = 2t_p$. The inverse of τ is the SW frequency f ($f = 1/\tau = 1/(2t_p)$). Typically, t_p ranges from 0.5 to 100 ms. ΔE represents the step potential of the staircase waveform. The opposing directions of the neighboring potential pulses induce the studied electrode reaction in both the anodic and cathodic directions, providing insights into the redox mechanism, similar to cyclic voltammetry. With respect to the overall potential modulation direction (panel A: E_s and E_f referring to the starting and final potential, respectively), forward and backward (reverse) potential pulses can be identified. Current is sampled at the end of each potential pulse (mean current of the last 5% of the current samples, for example) to discriminate against the charging current, resulting in a significant signal-to-noise ratio. **(C)** A typical SW voltammogram comprises forward current (I_f), reverse current (I_r), and net current (I_{net}) components. The forward and backward currents correspond to the current sampled at each forward and backward pulse, respectively (panel B), while the net component is the difference between them, $I_{net} = I_f - I_b$. All current components are plotted against the mid-potential of each potential cycle (i.e., the potential of the staircase ramp; panel A).

In addition to its analytical applications, when square-wave voltammetry (SWV) is employed for the fundamental study of electrode processes, the processing of voltammetric data and the comparison or fitting of theoretical and experimental data become crucial. The theoretical framework, specific to electrode mechanisms, is typically generalized by normalizing the current to a dimensionless form, which differs from the experimentally measured real current. This discrepancy makes the correlation between theory and experiment not straightforward. Another challenge in experimental analysis is the influence of numerous parameters that are difficult to precisely control, thus hindering the estimation of the real current in theoretical models. A typical example is the active surface area of a solid electrode, which may vary between experiments conducted with the same electrode. In theoretical models, the electrode surface area is typically neglected, as it is considered part of the parameters used for normalizing the current to a dimensionless form. Consequently, direct fitting of theoretical and experimental data is not possible. This highlights the need for appropriate processing of experimental data to uncover the underlying electrode mechanism and extract essential kinetic and thermodynamic parameters of the electrode reaction. The objective of this study is to provide a few simple guidelines to address this common issue in voltammetry, focusing on square-wave voltammetry (SWV) and its advanced variation, double-sampled square-wave voltammetry (DSSWV) [14].

RESULTS AND DISCUSSION

We consider a one-electron, quasi-reversible electrode reaction (1) involving a dissolved redox couple Red(sol)/Ox(sol) at a macroscopic planar electrode. The mathematical modeling of this reaction has been described elsewhere [3].

$$\mathrm{Red(sol)} \rightleftarrows \mathrm{Ox(sol)} + e^- \tag{1}$$

The real current I can be calculated as the product of Ψ, a dimensionless function derived solely through numerical simulations [15], and A, the amperometric constant. A is defined as $A = FSc(Df)^{1/2}$, where F is the Faraday constant, S is the electrode surface area, D is the common diffusion coefficient of both Red and Ox forms, c is the bulk concentration of the initially present reactant Red(sol), and f is the SW frequency. In general, the dimensionless function Ψ encompasses various aspects of the electrode mechanism, including the mass transfer regime, electrode kinetics, and other relevant phenomena like adsorption equilibria and coupled chemical reactions. This complexity makes the theory of voltammetry intricate in a broad context [3, 5, 6]. The role of theoretical analysis and mathematical modeling is to unveil the behavior of Ψ as a function of intrinsic kinetic parameters (such as the standard rate constant k_s and the electron transfer coefficient α, in the case of interfacial electron transfer modeled by the Butler-Volmer kinetic model [16]) and parameters related to the potential modulation (such as the SW frequency, amplitude (E_{sw}), and step potential (ΔE); see Fig. 1). For a single electrode reaction described by equation (1) and characterized by specific values of k_s, α, and D, when studied under a particular set of potential parameters (f, E_{sw}, and ΔE), the dimensionless function Ψ has a unique value. However, the actual current I cannot be calculated theoretically because it also depends on the amperometric constant A, which is often unknown due to the difficulty in precisely determining the electrode surface area S. The characteristics of the dimensionless function Ψ for the electrode reaction (1) are primarily governed by the dimensionless electrode kinetic parameter $\kappa = k_s/(Df)^{1/2}$ [3, 5, 6]. This parameter combines the effects of electrode kinetics (k_s), the rate of diffusion mass transfer (D), and the critical time of the voltammetric experiment (f). To examine the impact of electrode kinetics on the intensity of the dimensionless voltammetric response (specifically, the dimensionless net-peak current $\Delta\Psi_p$), one can vary the frequency of the potential modulation, thereby altering the critical kinetic parameter κ. A typical result of such theoretical analysis is presented in the inset of Figure 2.

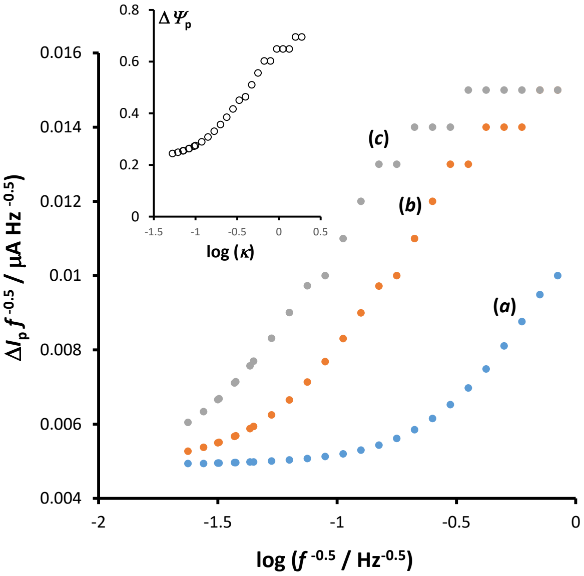


Figure 2. The figure illustrates the typical relationship between the frequency-normalized net peak current ($\Delta I_p/f^{1/2}$) and the logarithm of the inverse square root of the square-wave (SW) frequency, for three different standard rate constants: k_s = 0.001 (a), 0.005 (b), and 0.01 cm/s (c). The simulations were conducted under the following conditions: stoichiometric number of electrons in the electrode reaction equation $n = 1$, electron transfer coefficient $\alpha = 0.5$, common diffusion coefficient $D = 5 \times 10^{-6}$ cm^2/s, bulk concentration of the reactant $c = 1 \times 10^{-7}$ mol/cm^3, electrode surface area S = 0.001 cm^2, SW amplitude E_{sw} = 50 mV, step potential ΔE = 10 mV, and temperature T = 298.15 K. The inset graph presents the dependence of the dimensionless net peak current ($\Delta\Psi_p$) on the logarithm of the electrode kinetic parameter $\kappa = k_s/(Df)^{1/2}$, achieved by varying the SW frequency for a standard rate constant k_s = 0.005 cm/s. The other conditions remain the same as in the main panel.

Considering the actual current I in the same frequency analysis, analogous to a real experiment, one realizes that the effect of frequency is more complex, as it simultaneously affects both Ψ and A. To overcome this challenge, the real net peak current (ΔI_{p}) needs to be normalized with the corresponding frequency, i.e., $\Delta I_{\mathrm{p}}/f^{1/2}$. The function $\Delta I_{\mathrm{p}}/f^{1/2}$ vs. $1/f^{1/2}$, which qualitatively corresponds to the theoretical function $\Delta\Psi_{\mathrm{p}}$ vs. κ, is illustrated in Figure 2 for three different values of the standard rate constant. Both functions, $\Delta I_{\mathrm{p}}/f^{1/2}$ vs. $1/f^{1/2}$ and $\Delta\Psi_{\mathrm{p}}$ vs. κ, can be used to extract the standard rate constant k_{s} of the studied electrode reaction. However, they cannot be directly fitted, as the real net peak current also depends on the remaining unknown parameters of the amperometric constant A.

In order to ensure a reliable and meaningful comparison between I and Ψ, it is necessary to transform the real current into a dimensionless form, despite the unknown amperometric constant. Drawing inspiration from spectroscopy [17], a straightforward approach is to employ a self-normalization procedure where both the real current and the dimensionless current are normalized using the corresponding net-peak current. Specifically, the ratio $I/\Delta I_{\mathrm{p}}$ is equivalent to the ratio $\Psi/\Delta\Psi_{\mathrm{p}}$, given that $\Delta I_{\mathrm{p}} = \Delta\Psi_{\mathrm{p}}\,A$. This simple normalization enables direct comparison and fitting procedures, effectively eliminating the influence of the unknown amperometric constant. Consequently, a thorough analysis of the complete SW voltammogram can be conducted, taking into account all three voltammetric components: forward, backward, and net components. Figure 3 illustrates self-normalized SW voltammograms for three different values of the SW frequency corresponding to a typically quasireversible electrode reaction. The increase in frequency, ranging from very low (f = 1.4 Hz; Fig. 3a) to high values (f = 1000 Hz, Fig. 3c), causes the electrode reaction to transition from close-to-reversible to the characteristic electrochemically irreversible kinetic region [3, 5, 6]. This transition is evident from the morphological changes observed in the forward and backward voltammetric components. These dimensionless voltammograms can be effectively fitted with the theoretical function $\Psi/\Delta\Psi_{\mathrm{p}}$ to estimate the typical kinetic parameters of the electrode reaction, such as k_{s}, α, and D. An alternative approach to achieve full normalization of the real current is demonstrated in Figure 4. This approach examines the frequency-normalized real net-peak current ($\Delta I_{\mathrm{p}}/f^{1/2}$) as a function of frequency. The analysis in Figure 4 represents an improved version of the analysis depicted in Figure 2, as the ratio $\Delta I_{\mathrm{p}}/f^{1/2}$ is further normalized by dividing it by the value measured at the lowest frequency ($\Delta I_{\mathrm{p}}f^{-0.5}/\Delta I_{\mathrm{p}}f^{-0.5}(f_{\mathrm{min}})$). In the example presented in Figure 4, the minimum frequency is denoted as f_{min} = 1.413 Hz. The ratio $\Delta I_{\mathrm{p}}f^{-0.5}/\Delta I_{\mathrm{p}}f^{-0.5}(f_{\mathrm{min}})$ is dimensionless and equivalent to the ratio $\Delta\Psi_{\mathrm{p}}(f)/\Delta\Psi_{\mathrm{p}}(f_{\mathrm{min}})$, where $\Delta\Psi_{\mathrm{p}}(f)$ represents the dimensionless net-peak current simulated for a given frequency, and $\Delta\Psi_{\mathrm{p}}(f_{\mathrm{min}})$ is the dimensionless net-peak current corresponding to the first, minimum frequency within the selected frequency interval. Notably, Figure 4 demonstrates that the function $\Delta I_{\mathrm{p}}f^{-0.5}/\Delta I_{\mathrm{p}}f^{-0.5}(f_{\mathrm{min}})$ exhibits sensitivity to the standard rate constant. Consequently, this function can be directly fitted with simulated data to facilitate the kinetic characterization of the investigated electrode reaction.

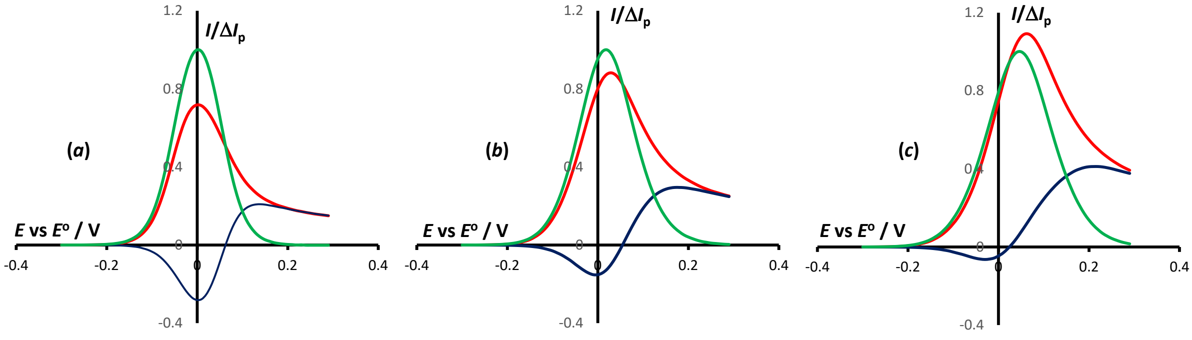


Figure 3. Representation of self-normalized SW voltammograms, illustrating the net (green), forward (red), and backward (blue) voltammetric components simulated for three different SW frequencies: f = 1.4 Hz (a), 251.2 Hz (b), and 1000 Hz (c). The standard rate constant is set to k_{s} = 0.001 cm s^{-1}. Self-normalization is achieved by dividing the current by the corresponding net peak-current (ΔI_{p}) of the voltammogram. All other conditions remain the same as in Figure 2.

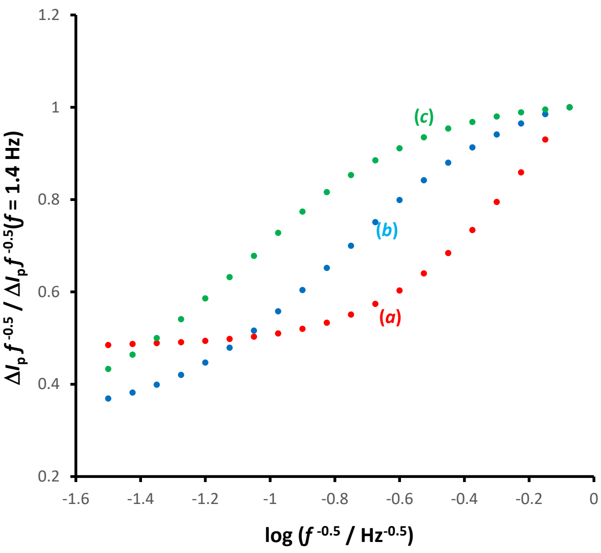
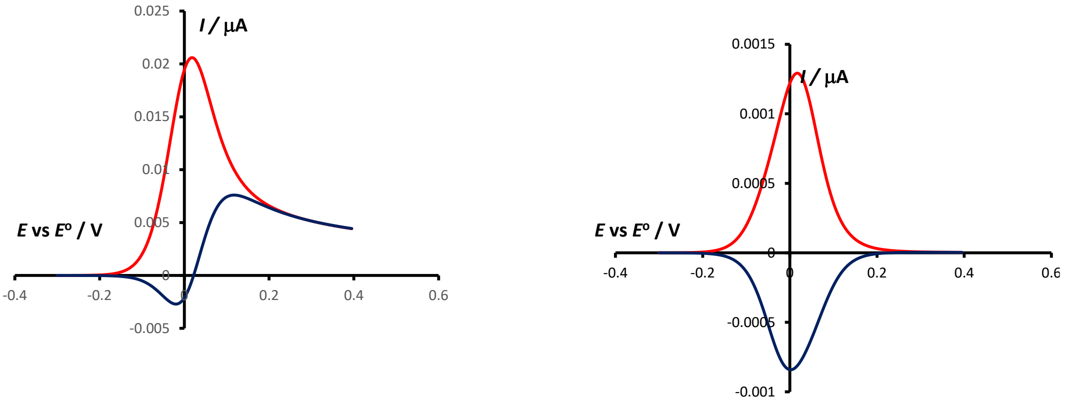


Fig. 4. The typical relationship between the frequency-normalized real net peak-current ($\Delta I_D/f^{1/2}$) and the logarithm of the inverse square root of the frequency for different standard rate constants: ks = 0.001 cm s-1 (a), 0.005 cm s-1 (b), and 0.01 cm s^{-1} (c). Furthermore, an additional normalization step is performed by dividing all values by the corresponding value obtained at the minimum frequency, denoted as ($\Delta I_D f^{-1/2}/\Delta I_D f^{-1/2}$ (f = 1.477 Hz)). It is worth noting that all other conditions remain the same as depicted in Figure 2.

In this section, we will discuss an advanced approach to process voltammetric data, focusing on a recently introduced technique called double-sampled square-wave voltammetry (DSSWV) [14].

The motivation behind developing DSSWV as an enhanced version of conventional SWV stems from the need for a comprehensive analysis of both forward and reverse components of the SW voltammogram when investigating the mechanistic and kinetic aspects of an electrode process. However, the backward component of the response often exhibits poor development, either due to sluggish electron transfer or the low amplitude of the SW pulses, as illustrated in Figure 5a. To overcome this limitation and broaden the applicability of SWV, a double-sampling current protocol was proposed. In this protocol, the current is sampled twice in the last quarter of the pulse duration, at designated times ts and tp (refer to Figure 1 in [14]). Subsequently, the current corresponding to each pulse is transformed into a differential current calculated as $I_{dif} = I(t_s) - I(t_p)$. The implications of this simple transformation are remarkable for both the forward and backward SW components, as demonstrated in Figure 5b. It is anticipated that this transformation of the SWV current components will enable improved mechanistic and kinetic analysis of electrode processes across various degrees of electrochemical reversibility. Furthermore, based on existing literature, there is a plausible expectation that DSSWV will enhance the analytical performance of the technique as well [18].



(a) **(b)**

Fig. 5. A comparison of conventional (a) and double-sampled square-wave voltammograms (b), illustrating the forward (red) and backward (blue) voltammetric components. The simulations are conducted for a standard rate constant of k_s = 0.005 cm s^{-1}, SW frequency of f = 10 Hz, SW amplitude E_{sw} = 25 mV, and step potential of ΔE = 5 mV. In the case of double-sampled square-wave voltammetry (DSSWV), the ratio of the sampling time to pulse duration is set at t_s/t_p = 0.8 [14]. All other conditions remain the same as depicted in Figure 2.

Since both forward and backward voltammetric components exhibit well-developed peaks regardless of the degree of electrochemical reversibility (as shown in Figure 5b), they serve as a foundation for rigorous kinetic analysis. In addition to the conventional approach of analyzing the relationship between their peak currents and peak potentials, it is valuable to consider all current values of both components to explore their detailed interdependence. Essentially, one can examine the characteristics of the function I_b vs. I_f within a specific voltammogram under various conditions of the electrode reaction. To achieve this, Figure 6 displays the dimensionless function $I_b/\Delta I_p$ vs. $I_f/\Delta I_p$, where ΔI_p represents the corresponding net-peak current of the DSSW voltammogram. This dimensionless representation allows for self-normalization, as explained earlier in the context of conventional SWV. By analyzing this function, one can gain insights into the interrelationship between the forward and backward voltammetric components and study their behavior under different conditions of the electrode reaction. The function $I_b/\Delta I_p$ vs. $I_f/\Delta I_p$, which represents a current-current function, should not be confused with a voltammogram, which typically represents a current-potential function. However, this current-current function offers a novel approach for advanced analysis of voltammetric data. It provides valuable insights and exhibits high sensitivity to the standard rate constant, making it particularly useful for studying very fast electrode reactions characterized by a rate constant of $k_s = 0.1$ cm s^{-1} (as depicted by curve d in Figure 6). This approach expands the kinetic range of accessible standard rate constants, particularly in the direction of very fast and nearly reversible electrode reactions. This advantage sets it apart from conventional analysis methods used in SWV [3].

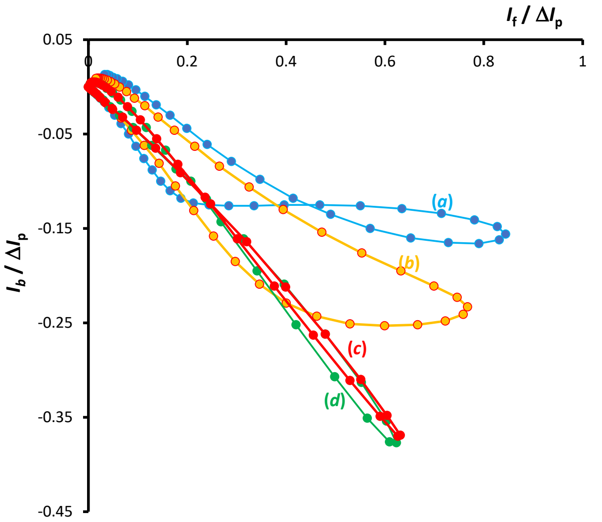


Fig. 6. Figure 6 displays the backward vs. forward voltammetric components of double-sampled square-wave voltammetry (DSSWV) for different standard rate constants: k_s = 0.005 cm s^{-1} (a), 0.01 cm s^{-1} (b), 0.05 cm s^{-1} (c), and 0.1 cm s^{-1} (d). The current values are self-normalized by dividing them by the corresponding net peak-current (ΔI_p) of the DSSW voltammogram. The simulations are conducted under the following conditions: SW frequency f = 100 Hz, SW amplitude E_{sw} = 25 mV, step potential ΔE = 10 mV, and a ratio of the sampling time to pulse duration t_s/t_p = 0.8. All other conditions remain the same as depicted in Figure 2.

It is worth noting that the current-current function depicted in Figure 6 can be mathematically interpreted within the context of complex numbers [19]. The forward and backward currents can be viewed as the real and imaginary components, respectively, of a complex current denoted as $Z = \Psi_f + i\Psi_b$. Here, Ψ_f represents the real part, Ψ_b represents the imaginary part, and $i = \sqrt{(-1)}$ is the imaginary unit. In order to study the behavior of Z, it is common to plot the imaginary component (Ψ_b) against the real component (Ψ_f). Therefore, the plot presented in Figure 6 represents the evolution of the complex number Z during the course of the voltammetric experiment, showcasing the relationship between Ψ_b and Ψ_f. Treating voltammetric data within the framework of complex numbers introduces a new approach for analysis, providing additional tools for advanced mechanistic and kinetic analysis of electrode processes. This perspective enhances the capabilities of voltammetric techniques and will be further elaborated upon in our future communications.

CONCLUSIONS

Accurate processing of electric current data obtained from a voltammetric experiment is essential for reliable comparison with theoretical predictions and understanding the mechanistic and kinetic aspects of an electrode process. The theory establishes a clear relationship between the experimentally measured current I and the current function Ψ, which can only be determined through numerical simulations. The relationship is defined as $I = nFSc(Df)^{1/2}\Psi$, where n represents the stoichiometric number of electrons involved, and other symbols are defined as explained in the previous section. To overcome any discrepancies between I and Ψ, a straightforward approach is to apply a self-normalization procedure to both currents using their corresponding net peak-currents. By dividing the currents by their respective net peak-currents, the functions $I/\Delta I_p$ and $\Psi/\Delta\Psi_p$ can be directly compared and fitted. This approach opens up new possibilities for analyzing voltammetric data, where the conventional current-potential function in SWV can be enriched with a novel current-current function. This new function utilizes the forward and backward components of the SW voltammogram, specifically the $I_f/\Delta I_p$ vs. $I_b/\Delta I_p$ function. The current-current function represents a promising and innovative tool for more in-depth and advanced processing of voltammetric data. It enables the expansion of the range of accessible rate constants, as demonstrated in the case of DSSWV.

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НАПРЕДНА ОБРАБОТКА НА ЕЛЕКТРОХЕМИСКИТЕ ПОДАТОЦИ ВО КВАДРАТНО-БРАНОВА ВОЛТАМЕТРИЈА

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Обработката на електрохемиските податоци во волтаметријата е од суштинска важност за изучување на реакциските механизми и кинетиката на електродните реакции. Квадратно-брановата волтаметрија е една од најнапредните пулсни волтаметриски техники која овозможува изучување на реакциските механизми, мерење брзи процеси на пренос на електрони, како и квантитативни определувања на аналити во субмикромоларно концентрациско подрачје. Заради релевантна споредба на експерименталните со теоретските податоци, во овој труд се предложени неколку нови идеи за обработка на електрохемиските податоците, земајќи ја предвид разликата во прикажување на реалната експериментална струја и бездимензионалната функција на струјата која се користи во теоретското моделирање на електродните процеси во квадратно-брановата волтаметрија. Важно е да се истакне дека во рамките на скорешно воведената нова техника наречена квадратно-бранова волтаметрија со двојно мерење на струјата, предложена е целосно нова методологија за анализа на податоците. Имено, вообичаената функција *струја-потенцијал* (позната како волтамограм) се трансформира во функција *струја-струја*, која има своја аналогија со комплексните броеви во математиката. Со примена на оваа методологија е проширен е интервалот на мерливи брзини на пренос на електрони, при што може да се мери кинетиката на многу брзи процеси на пренос на електрони (практично реверзибилни електродни реакции) кои се одликуваат со стандардна константа на брзина на пренос на електрони од редот на 0.1 cm s^{-1}.

Клучни зборови: квадратно-бранова волтаметрија; симулации; обработка на електрохемиски податоци; електродна кинетика; квадратно-бранова волтаметрија со двојно мерење на струјата

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Original scientific paper

NICKEL BASED METAL CARBIDES AS ELECTROCATALYSTS FOR METHANOL OXIDATION REACTION

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Effective catalysts play a crucial role in enhancing methanol oxidation within in cells. In light of this, diverse catalysts have undergone examination; however, recently, significant research interest has turned towards Ni-based metal carbides. Their appeal lies in their cost-effectiveness, high activity, and notably, the presence of metal-carbon bonds. These bonds are formed due to carbon's *s*- and *p*-hybridization with the metal d orbitals. This combination heightens the *d*-band activity of the metal, akin to that of Pt, as suggested by density functional theory (DFT) approximations. The catalytic potential can be further augmented through the combination of metal carbides with other active materials. Furthermore, refining the catalyst's size and morphology has the potential to enhance its electrochemical application for methanol oxidation. This involves maximizing the active surface area by optimizing its morphology and increasing the availability of metal carbide atoms. The introduction of multiple metals to metal carbides or the incorporation of multi-metal-based electrocatalysts onto the metal carbide's surface is also an area of fervent exploration. Such endeavors hold the promise of enhancing activity and devising economically viable methods for methanol oxidation-based fuel cells.

Key words: Nickel; metal carbide; MOR; fuel cells; Energy; Zero carbon emission

INTRODUCTION

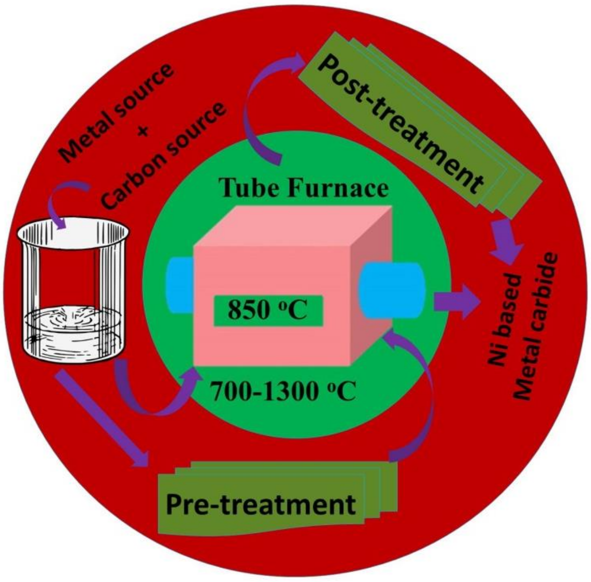
The primary energy source of current society is non-renewable fossil fuel, and its excessive consumption has led to energy and environmental problems. To mitigate these issues, an effective plan for environmentally friendly technology with renewable energy is needed [1–3]. Chemical energy conversion to electrical energy through fuel cells is the best possible option, considering its minimal or low environmental impact [4–6]. Among the available technologies, direct methanol fuel cells (DMFCs) are hailed as one of the most promising technologies, with methanol acting as the chemical fuel. Methanol offers various advantages, such as ease of transport and storage, low cost, low operating temperature, and high energy density [7–11]. However, DMFCs face two serious problems that limit their performance for practical applications: 1) the slow kinetics of electron transfer during the anodic oxidation of methanol, which affects the production rate of protons and consequently impacts power generation efficiency; 2) crossover of methanol, which is related to the electrolyte's catharsis ability [12–14]. DMFCs require an electrocatalyst to reduce the activation energy of the methanol oxidation reaction (MOR). So far, platinum (Pt) is the state-of-the-art electrocatalyst for MOR, but its poisoning effect caused by CO chemisorption significantly restricts its practical application [15, 16]. Although extensive research has been conducted to overcome this issue by combining it with other elements like PtRu, PtZn [17, 18], or two or more elements like PtRuIr, PtRuIrSn [19, 20], unfortunately, high cost, low availability, and poor stability still hinder their practical application [20–22].

Therefore, extensive investigative studies have been conducted to develop non-precious metal-based electrocatalysts for MOR [23]. Among these non-precious metals, nickel (Ni) has gained special research attention due to its low price, environmental friendliness, and, most importantly, its excellent surface oxidation properties [24, 25]. However, the catalytic application of Ni-based electrocatalysts faces challenges due to their poor electrical conductivity, low surface area, and limited durability. Consequently, the study of Ni-based MOR still offers unexplored opportunities for investigation [26].

To enhance the electrocatalyst's performance, morphology plays a vital role. For instance, one-dimensional (1D) electrocatalysts have demonstrated excellence in fast electron transportation, remarkable durability, and a high exposure of active lattice planes compared to their bulk counterparts [27–30]. Similarly, three-dimensional (3D) morphology retains the fundamental characteristics of bulk materials while offering a large surface area, excellent mechanical and chemical strength, high flexibility, an open and porous structure, and resistance to aggregation [31–33]. Additionally, 3D morphology boasts advantages such as abundant active sites, a high ratio for utilization, excellent electron/mass transfer kinetics, and impressive MOR activity coupled with outstanding stability and durability [34, 35].

Furthermore, two-dimensional (2D) materials have attracted extensive research attention owing to their shortened charge migration pathways, large surface area, and rich diffusion channels for molecules and ions. Prominent examples of 2D materials include transition metal dichalcogenides, layered double hydroxides, and MXenes, where MXenes encompass metal nitrides and carbides [36–38]. Intriguingly, these materials can alleviate stress resulting from volume changes and bubble generation due to surface reactions, resulting in excellent stability and activity.

Additional engineering approaches that can enhance catalyst performance encompass factors like thickness, porosity, size, doping, defects, crystal phases, and the preferential growth of active lattice planes [39]. Electrochemical applications can be further enhanced through the incorporation of carbon as a carbide with Ni or the introduction of co-catalysts alongside Ni. The incorporation of carbon with nickel enhances its catalytic performance due to improved conductivity, altered electronic configurations, and the presence of a large electronic cloud, particularly at their junction points. Co-catalysts like tungsten carbide (WC) can also enhance performance by providing excellent stability under thermal and electrochemical oxidation [40]. These co-catalysts serve both as catalysts for MOR oxidation and as supports to prevent the agglomeration of active materials, thus offering ample active sites for reactions [41, 42].



Scheme 1. A general diagram for the synthesis of Ni based metal carbide

The primary focus of this review article is to explore Ni carbides and other Ni-based metal carbides employed for methanol electrooxidation reactions. The study concludes that the incorporation of multi-metal concepts with synergistic effects has led to performance improvements. Additionally, the utilization of conductive supports has also enhanced performance. Morphology has played a significant role in boosting performance by offering a large surface area and active sites for reactions. A crucial consideration that merits further investigation is that Ni-based metal carbides possess the ability to facilitate MOR and produce formate instead of carbon dioxide/monoxide. The formate produced could find applications in various industries, and the superiority of these carbides over hydrogen oxidation has even been confirmed in some studies.

SYNTHETIC PROTOCOL

The synthetic route commonly employed for the synthesis of metal carbides involves the annealing method, as demonstrated in Scheme 1, with polyvinyl alcohol (PVA) being the most frequently utilized carbon source. In the annealing process, temperatures ranging from 700°C to 1300°C have been applied in various studies (Fig. 1). The primary rationale behind employing such high temperatures is the inherent stability of carbon bonds within the precursor materials. These bonds are notably robust and not easily broken under typical temperature conditions. Consequently, elevated temperatures are necessary to disrupt these sturdy bonds, initiating the desired reaction between carbon and metal. While the energy consumption associated with high temperatures is not ideal, the preparation of metal carbides often demands these conditions. Nevertheless, there are advantages to this method that can mitigate its drawbacks. The elevated temperature contributes to the formation of highly crystalline products and provides surface modifications that enhance the catalytic properties of the materials. Additionally, the high temperature facilitates the creation of a porous structure by releasing carbon dioxide and monoxide during the decomposition of reactants [43-45].

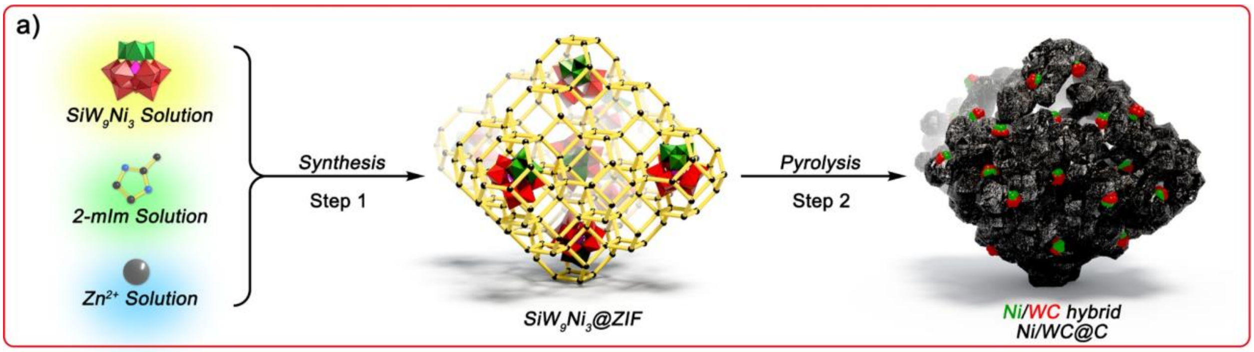


Fig. 1. Scheme for the synthesis of (a) Ni/WC Hybrid NPs (Ni/WC@C). Reprinted with permission from [45]. Copyright (2022) American Chemical Society

In tandem with the annealing method, some researchers have employed pre-treatments for materials. These pre-treatments can influence the composition or morphology of the final catalyst product. Notably, an intriguing procedure involves a one-pot solution-based strategy for synthesizing nickel carbide. In this approach, researchers dissolved nickel acetate in a mixture of oleylamine and Octadecene solvents. The solution was then subjected to a flux and reflux setup equipped with a magnetic stirrer. Employing a vacuum pump, low-boiling-point organic compounds, water, and air were removed from the solution at 80°C for 1 hour. Subsequently, the system was purged with argon gas and heated to 240°C at a rate of 5°C per minute. Heating was halted when a change in solution color was observed. The flask was then removed from the heating setup and allowed to cool to room temperature in a water bath. The resulting catalyst was collected and washed with ethanol to remove any unreacted reactants [46].

Electrochemical application

Fuel cells have demonstrated their potential as a promising energy source capable of addressing both energy and environmental concerns. Methanol serves as the primary fuel for these cells, with the oxidation of methanol yielding high energy output due to its remarkable energy density. Nevertheless, this process of methanol oxidation necessitates a catalyst to lower the activation energy required for oxidation. In this regard, numerous catalysts have undergone extensive study in pursuit of optimal performance. Among these catalysts, Ni-based metal carbides have garnered special research attention for various reasons, which will be extensively explained in the following lines. A comparative analysis for better understanding is presented in Table 1.

Among the array of catalysts investigated, Pt/Ni-WC was synthesized by Wang et al. [44] using a microwave-assisted annealing method. This catalyst exhibited a nanoparticle morphology and showcased catalytic performance of 0.51 mA/cm^2 at 0.4 V vs Ag/AgCl in an electrolyte comprising 1.0 M CH_3OH and 0.5 M H_2SO_4. Notably, this catalyst exhibited superior stability compared to Pt/C, attributed to the robust interaction between Pt and Ni-WC, resulting in sustained durability over a span of 7000 seconds. Although this catalyst demonstrated activity in the methanol oxidation reaction, its response remained modest, falling short of meeting the societal energy demands. The intrinsic value of Pt, being a precious metal, further raises concerns among scientists regarding its practical application in fuel cells. Consequently, the pursuit of precious metal-free electrocatalysts with efficient performance has become a paramount objective. Urgent attention is required to enhance the efficiency of the methanol oxidation reaction, addressing the pressing need for higher energy output.

Comparatively, good work in this direction was achieved by Wang et al. [43], who introduced PtNiPb/WC in an electrolyte comprising 0.5 M CH_3OH and 0.5 M H_2SO_4. This catalyst exhibited a remarkable current density of 2.67 mA/cm^2 at an overpotential of 0.85 V vs RHE. This enhanced performance, surpassing the capabilities of previously discussed materials, can be attributed to the presence of multiple metals in the sample. The synergistic effects resulting from these multimetals within the catalyst contributed to its heightened performance. Impressively, the catalyst also demonstrated excellent stability over a prolonged duration of approximately 1.5 h. Despite this improvement, the achieved performance still falls short of effectively addressing the energy crisis. The inclusion of Pt within the catalyst remains a drawback due to its associated cost and availability limitations.

In a similar vein, Zhang et al. [48] utilized a microwave-assisted reduction approach to craft a Ni-MoC$_x$/C-supported Pt (10 wt%) electrocatalyst (10Pt/Ni-MoC$_x$/C), which exhibited pronounced electrocatalytic proficiency in methanol oxidation. The outcomes underscored the superior electrocatalytic activity and stability of the 10Pt/Ni-MoC$_x$/C electrocatalyst in comparison to the 20 wt% Pt/C (20Pt/C) alternative. Notably, the electrochemical surface area of 10Pt/Ni-MoC$_x$/C attained 68.4 m^2/g, surpassing 20Pt/C's 63.2 m^2/g. This heightened stability and activity within the 10Pt/Ni-MoC$_x$/C electrocatalyst can be ascribed to two key factors: firstly, the anchoring effect stemming from Ni and MoC$_x$ formation during the carbonthermal reduction process; and secondly, the synergistic interplay among Pt, Ni, MoOx, and MoCx. These findings underscored the promise of 10Pt/Ni-MoC$_x$/C as an effective electrocatalyst for direct methanol fuel cells.

Remarkably, the catalyst 10Pt/Ni-MoC$_x$/C exhibited a current density of 260.5 mA/mg at an overpotential of 0.9 V vs RHE in an electrolyte comprising 1.0 M CH_3OH and 0.5 M H_2SO_4. This exceptional performance can be attributed to the presence of Pt, along with the extensive surface area where nanoparticles are uniformly dispersed, preventing agglomeration and ensuring maximum active sites for reactions. The inclusion of Mo in the composition notably heightened the responsiveness to methanol oxidation. The synergistic interaction among multiple atoms (Ni, Mo, Pt) facilitated heightened electronic cloud density, particularly at their junctions, thus proving immensely effective in methanol oxidation reactions. Additionally, the presence of carbon as a support accelerated rapid electron transport, further amplifying this catalytic activity. While the material exhibits excellence and holds potential for practical applications, the presence of precious metals might impede industrialization due to their elevated costs and limited availability. Consequently, it is highly advisable to focus on developing electrocatalysts based on transition metals and their carbides, excluding precious metals, to overcome these limitations.

In recent times, non-precious catalysts with efficient performance have been extensively investigated. For instance, Hou et al. [49] developed a Ni-WC based catalyst on the surface of a carbon aerogel derived from bacterial cellulose. The catalyst exhibited a current density of 105.7 mA/cm^2 at an overpotential of 0.8 V vs SCE. The optimal reaction medium was found to be 0.5 M CH_3OH and 1.0 M KOH, showcasing excellent stability over 3600 s. Moreover, with the introduction of a novel electrolyte, the peak current density rebounds to 88.42% of the initial value, highlighting notable stability. The synergy between Ni and WC produces a collaborative effect that facilitates the formation of NiOOH, subsequently diminishing the surface adsorption of CO. This effect effectively mitigates the temporary blockage of active catalytic sites. The achieved W_1Ni_{10}/CA composite in this study, known for its affordability, substantial specific surface area, elevated catalytic efficiency, and impressive resistance to CO-induced inhibition, emerges as a promising catalyst for application in DMFC scenarios. The response is promising but still requires performance improvement, leaving room for further advancements in catalytic applications that require research.

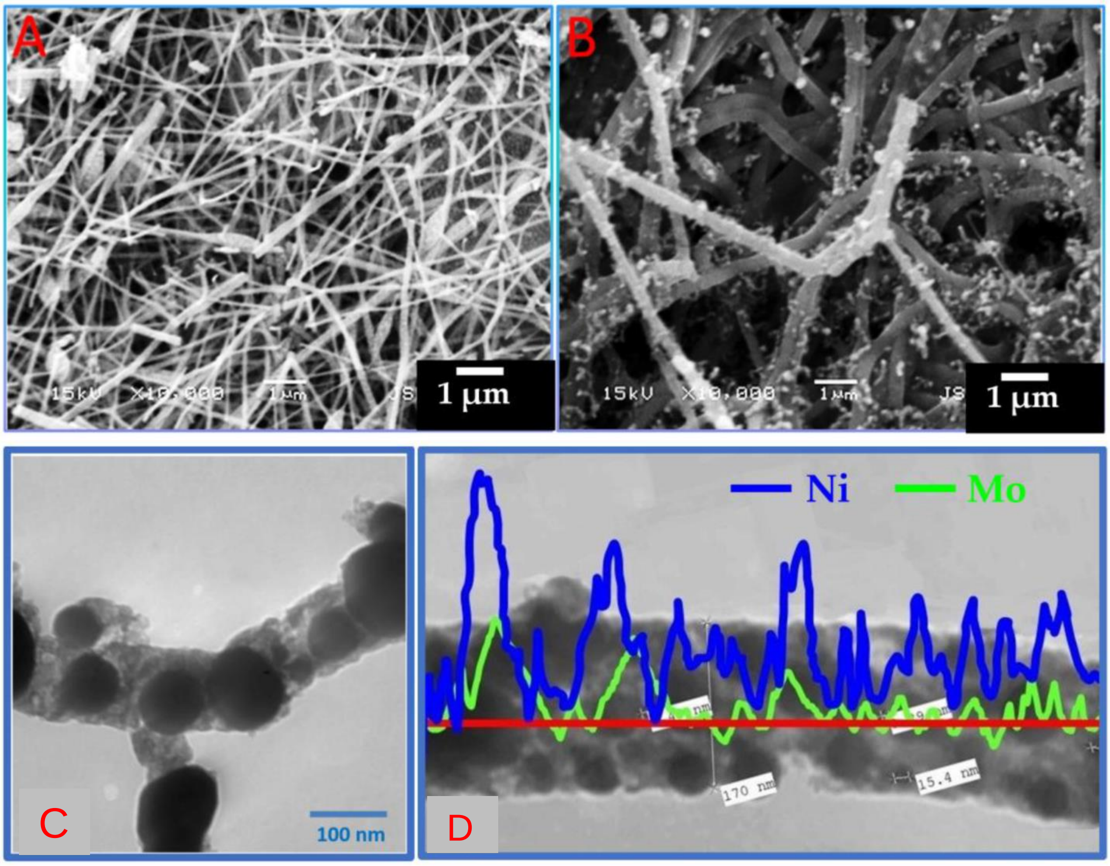


Fig. 2. SEM images of the produced Mo_2C–Ni–C composite nanofibers calcined at 850 °C and obtained from electrospun nanofibers containing 10 (A) and 35 (B) wt.% $MoCl_2$ compared to $Ni(AC)_2$. TEM image of Mo_2C/Ni/graphite composite nanofibers generated from 10% Mo/electrospun solution calcined at 850 °C (C) and linear elemental mapping for Ni and Mo along randomly selected (D). Reproduced with permission from Marwa M. Abdel-A, Polymer; published by MDPI, 2023 [47].

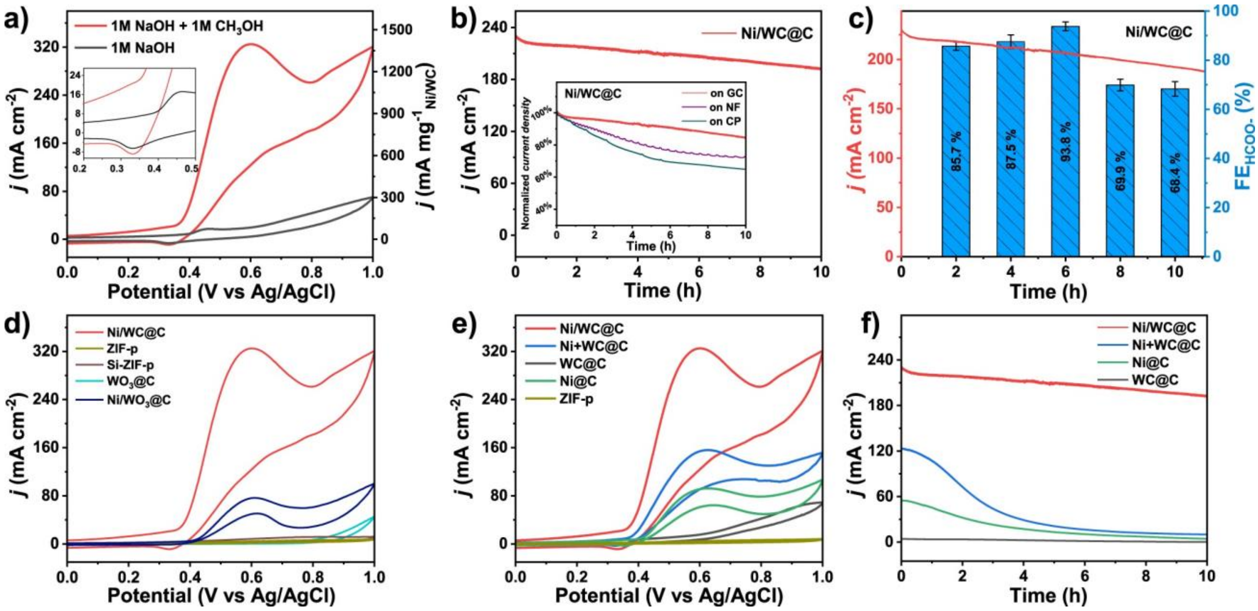


Fig. 3. (a) CV curves of Ni/WC@C in 1 M NaOH with and without 1 M methanol. (b) Chronoamperograms of Ni/WC@C in 1 M NaOH +1 M CH_3OH at 0.5 V versus Ag/AgCl/3.5 M KCl with a salt bridge. [Inset: chronoamperograms of Ni/WC@C when using GC (red), NF (purple),and CP (green) as substrate]. (c) Chronoamperograms (red line) and FE_{HCOO^-} (blue columns) of Ni/WC@C at 0.5 V versus Ag/AgCl/3.5 M KCl with a salt bridge. (d) CV curves of different samples for MOR in 1 M NaOH + 1 M CH_3OH. (e) CV curves of different catalysts for MOR in1 M NaOH + 1 M CH_3OH. (f) Chronoamperograms of different catalysts in 1 M NaOH + 1 M CH_3OH at 0.5 V versus Ag/AgCl/3.5 M KCl witha salt bridge. Reprinted with permission from [45]. Copyright (2022) American Chemical Society

Notably, the integration of Mo has demonstrated exceptional efficacy in electrocatalytic applications. For instance, Abdel-Aty and her team [47] developed an electrocatalyst, Mo_2C/Ni-CNFs (5%) for MOR, where the CNFs provide one-dimensional support for well dispersion of Mo_2C/Ni as shown in Fig. 2. The catalyst, necessitating a 1.0V overpotential vs Ag/AgCl to yield a current density of 107 mA/cm^2 in a 3.0 M CH_3OH and 1.0 M KOH electrolyte as shown in Fig. 4. This catalyst was synthesized through electrospinning a blend of poly(vinyl alcohol), nickel acetate, and molybdenum chloride, followed by vacuum calcination. This procedure resulted in the formation of zero-valent nickel and molybdenum carbide nanoparticles within amorphous graphite nanofibers due to the breakdown of metal constituents and polymer graphitization. The optimization of molybdenum content significantly contributed to achieving consistent and efficient electrocatalytic performance in methanol electrooxidation. For attaining the maximum methanol electrooxidation rate alongside electrode regeneration, employing a 5 wt.% molybdenum precursor relative to nickel acetate in the initial electrospun solution was identified as ideal. The calcination temperature played a pivotal role, notably enhancing methanol electrooxidation activity at 850°C. Employing Taguchi's robust design approach facilitated the identification of optimal conditions, resulting in the highest achievable current density. The parameters derived from this approach were as follows: 5 wt.% molybdenum content, 2.65 M methanol concentration, and a reaction temperature of 50°C. Interestingly, the reaction rate exhibited an inverse relationship with the medium temperature. The peak rate of methanol electrooxidation was attained at 45°C, while Taguchi's analysis pinpointed the optimum at 50°C. Given its remarkable performance, the proposed nanofibrous composite of Mo_2C/Ni-incorporated carbon emerges as a strongly recommended electrocatalyst for methanol oxidation, warranting further exploration of its potential with other organic substrates. The exceptional material response stems from the synergistic effects of multimetal interactions, particularly the bonding of Mo with carbon, leading to significant alterations in electronic configuration and the generation of a substantial electronic cloud at the junction. This electronic cloud was further augmented by the inclusion of Ni. Furthermore, the well-dispersion of active materials with minimum agglomeration and maximum active spots for reaction enhanced its application. The presence of carbon nanofibers, with their conductivity and one-dimensional structure, enhances electron transport and catalytic application of the final material. The catalytic application response can be further improved through morphological engineering and by providing a conductive support to enhance electron transport and achieve excellent MOR performance. In this context, Xie et al. [26] developed CNT-Ni/SiC, an electrocatalyst for MOR application. The system yielded an outstanding current density of 210 mA/cm^2 at an overpotential of 0.8 V vs Ag/AgCl in an electrolyte of 3.0 M CH_3OH and 1.0 M KOH. The catalyst exhibited exceptional durability for 4000 seconds continuously. The remarkable electrochemical performance of the composite stems from the uniform dispersion and secure anchoring of Ni nanoparticles, combined with the favorable electrical conductivity of the network-style framework established by CNTs and SiC. This three-dimensional structure offers a substantial surface area, optimizing reactive sites and minimizing diffusion paths. The synergistic effects of Ni and SiC amplify the electronic cloud at the junction, resulting in configuration changes that further enhance the response. Furthermore, the conductive surface and one-dimensional architecture accelerate electron transfer, leading to an enhanced response. Anticipations suggest that this cost-effective and practical synthesis method could be extended to create varied carbon-based and metallic hierarchical nanostructures with a wide range of applications.

Metal carbides open up another intriguing avenue that could enhance energy applications. Particularly, Ni-based metal carbides show promise in converting methanol into formate, exhibiting impressive performance and yielding high current densities. Moreover, this formate could be subjected to further oxidation to harness more energy or could find applications across diverse industries. In line with this, Li et al. [46] introduced Ni_3C as an electrocatalyst for methanol oxidation to formate, achieving a noteworthy current density of 127 mA/cm^2 at an overpotential of 0.6V vs Ag/AgCl. The catalyst also demonstrated stability over 5 hours in an electrolyte containing 1.0 M CH_3OH and 1.0 M KOH. This excellent performance arises from the strong nickel-carbon bonding, which enhances electron flow at their junction by modifying their configuration to facilitate the reaction. Furthermore, the morphology also plays a vital role: the material exhibits a nanostructure with nanoneedles that amplify surface area, diminish diffusion kinetics, and maximize active sites for reactions. This research is immensely valuable and merits further attention due to its exceptional energy response while emitting zero carbon.

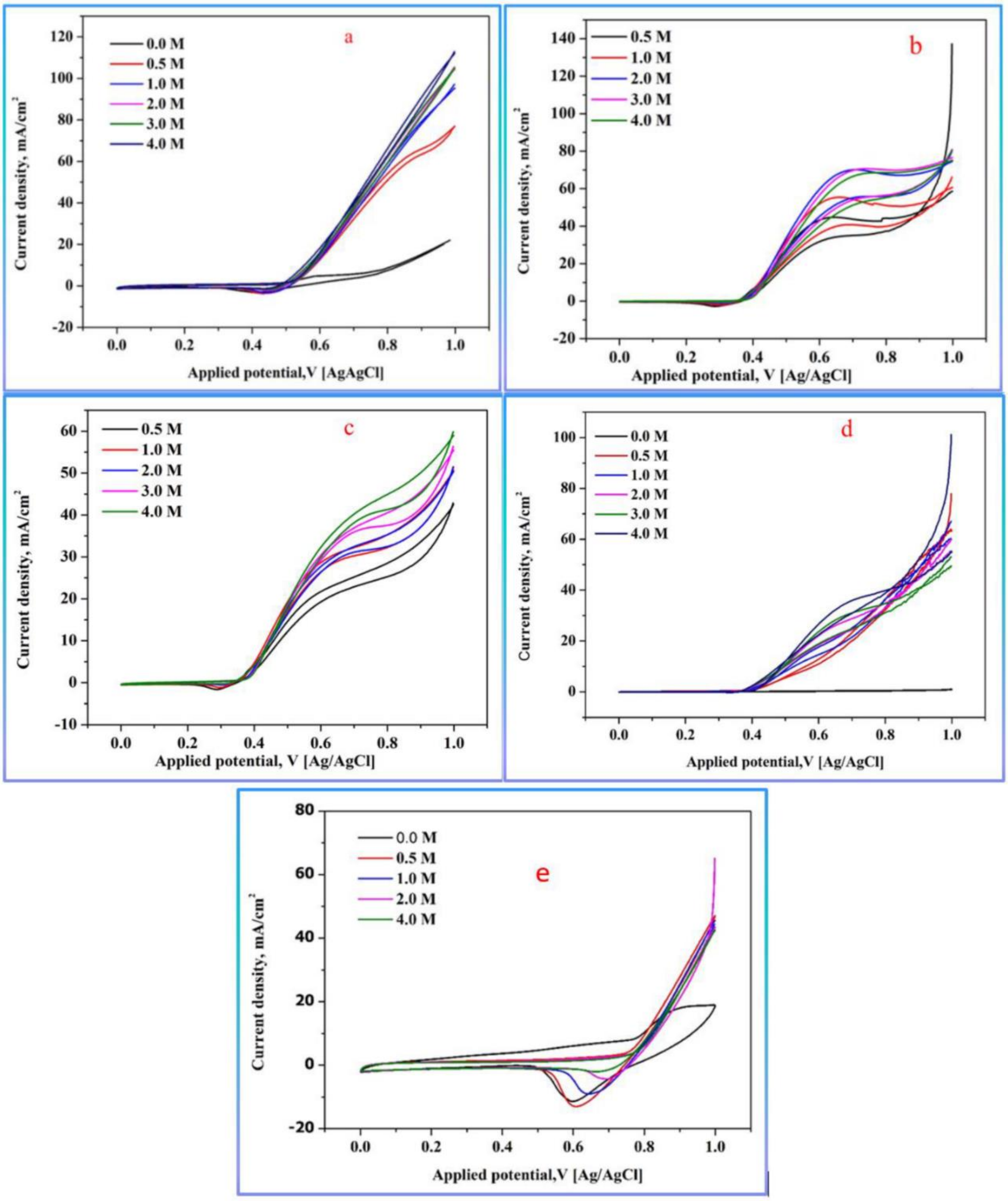


Fig. 4. Influence of molybdenum carbide content on the electrocatalytic activity of Mo_2C–NiCNFs prepared at 850 °C calcination temperature from electrospun solutions having different molybdenum chloride content: 5; (a), 10; (b), 15; (c), 25; (d) and 35 wt.%; (e), at different methanol concentration and scan rate of 50 mV/s. Reproduced with permission from Marwa M. Abdel-A, Polymer; published by MDPI, 2023 [47].

In contrast, other fuel cells that undergo complete methanol oxidation release carbon dioxide (albeit at low levels). This field requires additional exploration to enhance performance, whether through the introduction of multimetal catalysts, surface engineering, conductive support surfaces, and more. In this regard, Zhang et al. [45] employed the concept of multimetal synergy to achieve a high electronic cloud for exceptional MOR application. They prepared an electrocatalyst, Ni/WC, which required an overpotential of 0.6 V vs Ag/AgCl to achieve a current density of 325 mA/cm^2 in an electrolyte of 1 M CH_3OH and 1 M NaOH aqueous solution as shown in Fig. 3. The catalyst exhibited outstanding stability over a duration of 10 hours, confirming its robustness and practicality for MOR application in fuel cells.

The hybrid Ni/WC nanoparticles synthesized in this study display remarkable electrooxidation capabilities when employed for the oxidation of methanol, ethanol, iso-propanol, ethylene glycol, and propylene glycol in an alkaline solution. Particularly noteworthy in the context of methanol oxidation, the Ni/WC nanoparticles exhibit heightened reaction activity (with a mass activity of 1363 mA/mg) alongside exceptional stability (with only approximately a 6.8% drop in catalytic current after 4 hours). During the initial 6-hour electrolysis period, methanol efficiently transforms into formate with an approximate Faraday efficiency of 93.8%. Through the utilization of in situ IR spectra and control catalyst experiments, it has been established that both tungsten carbide (WC) and nickel oxyhydroxide (NiOOH, derived from Ni) species are capable of activating distinct functional groups within methanol. Importantly, their synergistic hybrid structure (Ni/WC) leads to an enhanced catalytic performance in converting methanol into formate. The catalyst's exceptional performance can be attributed to its nanosized particles, which provide an excellent large surface area for electrochemical reactions.

Table 1. Comparison of Ni based metal carbide MOR performance

S.No	Catalyst	Potential	Current density	Electrolyte	Ref
1	CNT–Ni/SiC	0.8 V vs Ag/AgCl	210 mA/cm^2	1.0 M CH_3OH 1.0 M KOH	[26]
2	PtNiPb/WC	0.85 V vs RHE	2.67 mA/cm^2	0.5 M CH_3OH 0.5 M H_2SO_4	[43]
3	Pt/Ni-WC	0.4 V vs Ag/AgCl	0.51 mA/cm^2	1.0 M CH_3OH 0.5 M H_2SO_4	[44]
4	Ni/WC	0.6 V vs Ag/AgCl	325 mA/cm^2	1.0 M CH_3OH 1.0 M NaOH	[45]
5	Ni_3C	0.6 V vs Ag/AgCl	127 mA/cm^2	1.0 M CH_3OH 1.0 M KOH	[46]
6	Mo_2C/Ni-CNFs (5%)	1.0 V vs Ag/AgCl	107 mA/cm^2	3.0 M CH_3OH 1.0 M KOH	[47]
7	10Pt/Ni-MoC*x*/C	0.9 V vs RHE	260.5 mA/mg	1.0 M CH_3OH 0.5 M H_2SO_4	[48]
8	Ni-WC	0.8 V vs SCE	105.7 mA/cm^2	0.5 M CH_3OH 1.0 M KOH	[49]
9	Ni/TaC	0.4 V vs SHE	1.1 mA/cm^2	--- 1.5 M H_2SO_4	[50]

It is highly interesting and needs more attention that Burstein et al. [50] conducted a study on metal carbides; the Ni/TaC catalyst displayed an excellent response to heated methanol when introduced to a chamber for catalytic applications. The system exhibited a current density of 1.1 mA/cm^2 at an overpotential of 0.4 V vs SHE in an electrolyte of 1.5 M H_2SO_4. This study demonstrates the superiority of methanol-based fuel cells over hydrogen oxidation reactions. The described electrocatalyst, composed of tantalum, nickel, and carbon, demonstrates higher catalytic activity in the anodic oxidation of methanol compared to hydrogen. This differentiation is attributed to the functionalization of surface carbon atoms through anodic oxidation, enabling them to adsorb and react with methanol. The proposed surface intermediate adopts a ring structure. This innovative mechanism for the anodic oxidation of methanol is exclusive to carbon-based surfaces and doesn't apply to metal electrocatalysts. Furthermore, it is specific to the anodic oxidation of carbon-based fuels and isn't relevant to hydrogen oxidation. The presence of the nickel and/or tantalum component is thought to enhance this observed behavior.

CONCLUSION AND FUTURE PERSPECTIVE

The growing concern over energy crises and environmental issues has prompted significant research endeavors toward the development of renewable energy sources as potential solutions. Among these, fuel cells emerge as a promising technology to address these challenges. Extensive research has been dedicated to exploring diverse electrocatalysts capable of efficiently oxidizing methanol to generate energy. Within this landscape, nickel-based metal carbide materials have garnered notable attention, attributed to their cost-effectiveness, efficiency, and suitability for electrochemical methanol oxidation reactions (MOR). Hence, the focus of this study is directed toward nickel-based metal carbides.

The heightened catalytic efficacy of metal carbides can be traced to the formation of metal-carbon bonds arising from the hybridization of carbon's *s*- and *p*-orbitals with the metal's *d*-orbitals, corroborated by DFT calculations. This hybridization leads to the expansion of the metal d band, rivaling that of Pt. Empirical evidence affirms that electrochemical MOR applications are positively correlated with diminishing catalyst size. Similarly, enhanced morphology facilitates optimal utilization of the expansive surface area and maximal incorporation of metal carbide atoms. Moreover, the catalytic impact can be further enhanced when metal carbides are amalgamated with other active materials. Incorporating doped and multimetal elements in electrocatalysts has demonstrated improved sensing activity.

To markedly enhance catalytic performance and achieve practically efficient electrocatalysts for MOR, it is recommended to reduce and control the catalyst size down to the nano or sub-nano scale. Counteracting nanoparticle agglomeration is pivotal to preserving a heightened active surface area and attaining elevated activity levels. In this context, various supporting materials, such as 3D graphene, boron nitride, graphitic carbon nitride, and carbon nanotubes, can be employed to mitigate nanoparticle agglomeration. These materials, in addition to curbing agglomeration, offer high conductive surfaces that expedite electron transport, further facilitating catalytic applications.

Morphology also exerts a substantial influence on catalytic activity. Transforming metal carbides into diverse morphologies, such as nanoflowers, nanoneedles, urchins, nanotubes, nanofibers, porous structures, and ultrathin layers, can amplify their activity. These unique morphologies confer extensive surface areas, enabling optimal utilization of active materials for targeted reactions while minimizing wastage of materials embedded deep within the catalyst's bulk structure. Additionally, the strategic integration of multiple metals within a single catalyst can augment activity by introducing a greater pool of active materials for catalytic processes. The observed synergistic effects in multimetal-based electrocatalysts contribute to their heightened activity levels as well.

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МЕТАЛНИ КАРБИДИ ШТО СОДРЖАТ НИКЕЛ КАКО КАТАЛИЗАТОРИ ЗА РЕАКЦИЈАТА НА ОКСИДАЦИЈА НА МЕТАНОЛОТ

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Катализаторите играат важна улога за подобрување на ефикасноста на процесот на оксидација на метанолот. Голем број катализатори се предмет на тековни истражувања, при што од неодамна постои значителен интерес за катализатори што содржат никел. Нивната соодветност произлегува од постоењето на метал-јаглеродна врска, како и од фактот дека тие се евтини и значително ефикасни. Метал-јаглеродната врска се формира меѓу *s*- и *p*-хибридните орбитали на јаглеродот и *d*-орбиталите на никелот, слично како кај платинските катализатори, според заклучоците на теоретските студии. Каталитичкиот ефект може да биде зголемен и преку комбинирање на металкарбидите со други активни метали. Од друга страна, морфологијата и големината на каталитичките честички имаат значајна улога. Притоа, особено е важно оптимизирањето на морфологијата на честичките во насока на поголема достапноста на супстратот до металкарбидната врска на катализаторот. Покрај тоа, особен интерес побудува комбинацијата на металкарбидите со честичките на различни метали зашто овозможува значително подобрување на каталитичките особини. Оваа целокупна област на истражување овозможува дизајнирање на ефикасни и евтини методи за развој на горивни ќелии што користат метанол.

Клучни зборови: никел, метал карбиди, горивни ќелии, енергија, нула-јаглеродна емисија

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