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Toward Smart Connected Health: IoT-Enabled Biosensors for Nicotine Metabolite Detection Using Nanomaterials and Machine Intelligence

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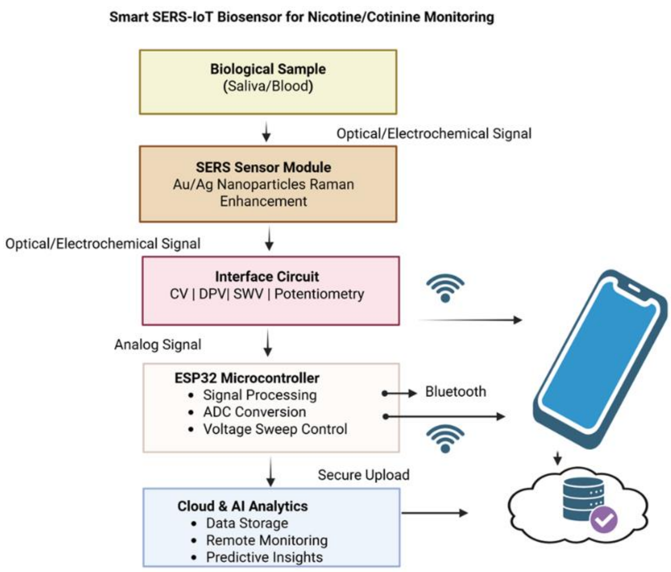
Biosensors; Nicotine; Cotinine; Nanomaterials; Electrochemical sensing; Artificial intelligence; Machine learning; Smartphone-based diagnostics; Point-of-care devices; Tobacco metabolite detection; Internet of Things (IoT)

ABSTRACT

One of the most concerning critical global health issues is tobacco consumption, which contributes significantly to preventable morbidity and mortality. Timely and accurate detection of cotinine, a primary metabolite of nicotine, is crucial for suggesting cessation programs. Gas chromatography-mass spectrometry, high-performance liquid chromatography, and immunoassays offer high sensitivity and specificity but are limited by high costs, the need for laboratory setup, and sophisticated techniques. The emergence of biosensors has led to the development of point-of-care (POC) alternatives due to their portability, ease of use, and cost-effectiveness. Their sensitivity and selectivity can be enhanced by the incorporation of nanomaterials, such as graphene, carbon nanotubes, and metal nanoparticles, enabling the detection of trace levels of nicotine metabolites in biological fluids, and the integration of AL and ML algorithms with the biosensors to process complex electrochemical signals, extract meaningful features, and quantify accurate analyte concentrations. In addition, the integration of biosensors with Internet of Things (IoT) infrastructure through smartphones and wireless modules such as Bluetooth, Wi-Fi, and NFC enables seamless real-time data acquisition, cloud-based storage, remote monitoring, and personalized health feedback. These types of biosensors, incorporated with AI & ML and IoT, offer the possibility of continuous, user-friendly, and connected monitoring of nicotine metabolite exposure, bridging the gap between laboratory diagnosis and digital health ecosystems. This review proposes a biosensor based on Surface Enhanced

Raman Spectroscopy (SERS) for the detection of nicotine metabolites in biological samples using Au/Ag nanoparticles. The Biosensor is integrated with a smartphone via Bluetooth, NFC, and Wi-Fi using an ESP32 Microcontroller, leading to seamless telemonitoring. The role of AI/ML in enhancing signal interpretation and the transformative impact of IoT integration in enabling connected health systems. It also discusses the current challenges related to reproducibility, stability, data security, and regulatory compliance, and outlines future directions for the development of smart biosensing technologies for precision tobacco exposure monitoring.



1. INTRODUCTION

Nicotine (IUPAC name: 3-[(2S)-1-methylpyrrolidin-2-ylpyridine], NIC) dependence is a leading cause of morbidity and mortality worldwide, associated with increased risks of cancer, cardiovascular disease, and pulmonary disorders [1]. Accurate monitoring of nicotine exposure is critical for clinical diagnostics, public health surveillance, and smoking cessation interventions. Cotinine, the primary metabolite of nicotine, serves as a robust biomarker due to its longer half-life [2]. One key point to consider is that tobacco smoke still lacks a perfect measurement method. Accurate measurement is essential for validating risk assessments related to tobacco exposure. Historically, cotinine has been measured from samples of salive, urine and boold. While traditional methods like gas chromatography–mass spectrometry (GC–MS) and enzyme-linked immunoassays (ELISA) provide high accuracy, they involve long processing times, high costs, and require laboratory infrastructure [3]. As a result, there is an increasing need for low-cost, portable, and real-time monitoring tools. Recent developments in nanomaterials and biosensor technology offer promising alternatives. Nanostructures, such as graphene, carbon nanotubes, and noble metal nanoparticles, exhibit superior electrical, optical, and catalytic properties, making them ideal transducers for biosensing [4]. Moreover, the integration of AI and machine learning (ML) enables efficient bio-signal analysis, enhancing sensor selectivity and accuracy. Biomarker identification using Machine Learning was aided by Artificial Intelligence for classification and Clustering, facilitating comprehensive molecular profiling analysis. By employing top-down approaches, such as Systematic Evolution of Ligands by Exponential Enrichment (S ELEX), the AI accelerates the discovery of high-specificity bioreceptors. In data analysis, Machine Learning models process complex datasets to classify, predict, and interpret biomolecular interactions with high accuracy. [5]. This review proposes a smart biosensor platform that combines nanomaterial-based transduction with AI-assisted data processing for nicotine metabolite detection

2. METHODS

Data was collected from various sources such as Elsevier, Google Scholar, IEEE, and the American Chemical Society (ACS) by searching keywords like "Biosensors", "Nicotine", Cotinine", "Nanomaterials", "Electrochemical sensing", "Artificial intelligence", "Machine learning", "Smartphone-based diagnostics", "Point-of-care devices", "Tobacco metabolite detection", "Internet of Things" all the diagrams were drawn using Microsoft PowerPoint version 2580 and Biorender.

3. CURRENT BIOSENSING TECHNOLOGIES FOR NICOTINE METABOLITES:

3.1Electrochemical Sensors:

Traditional techniques like High performance liquid chromatography (HPLC), Gas chromatography ma ss-spectroscopy (GC-MS) [6]. High performance liquid chromatography mass-spectroscopy (HPLC-MS), Gas chromatography (GC) [7] were used for the nicotine detection, which were very sophisticated, time-consuming, and required preliminary extraction and purification of NIC sample, and involved many steps. Now, researchers are switching to electrochemical sensors because of their fast response, relatively high sensitivity, low detection capabilities, and simple, low-cost, and reliable [8]. Shehata et al. developed an electrochemical sensor for NIC detection. Nano-TiO_2 wi th a carbon paste electrode was used to construct the sensor, and NIC was detected using a Nano-TiO_2 modified carbon paste sensor (NTMCP) within the range of 2×10^{-6} M to 5.4×10^{-4} M using differential pulse voltammetry. With a limit of detection of 1.34×10^{-8} M, the sensor demonstrates high sensitivity, simplicity, and selectivity for determining NIC levels in urine, which were compared to real cigarettes. Another study conducted by Li et al. involved synthesizing nitrog en-doped graphene sheets (NGS) and characterizing them using Transmission Electron Microscopy (TEM), Raman Spectroscopy, X-Ray Diffraction (XRD), and X-ray photoelectron spectroscopy (XPS). These nitrogen-doped graphene sheets were used to modify screen-printed carbon electrodes (SPCE). Li et al. demonstrated a detection sensitivity of 0.627 mA·cm− 2 mM− 1 with a limit of detection at 47 nM NIC, illustrating the sensors' sensitivity as shown by Li et al. Paper-based potentiometric sensors have also been developed. Electrodes were based on nicotinium cation (Nic) with either tetraphenylborate (TPB) or 5-nitro barbiturate (NB) counter anions as sensing materials for NIC recognition. It has been shown that the s ensors showed rapid and stable response with the detection limits of 6.0 and 8.0 µM for [Nic/TPB] and [Nic/NB], respectively, and showed that the sensors revealed a constant response over 3.5-6.5 pH. This design of the device makes it a portable, inexpensive way to detect trace levels of nicotine. NIC levels from human sweat from heavy smokers and from different types of cigarettes were measured, and the obtained results were compared favourably with results obtained from gas chromatography [9]. In a study done by Dayan et al., there is a proposal for a 3D-printed electrochemical sensor, which was printed using a 3D-printing pen, that quantified NIC levels from e-cigarettes and sweat samples. LOD was 2.7 mgL−1, without any electrochemical pre-treatment, showing its efficiency in direct detection and low cost. Au tadpole-like nanostructures immobilized onto titanium oxide (TiO) were synthesised and characterized using transmission electron microscopy (TEM), X-ray Diffraction (XRD), and X-ray photoelectron spectroscopy (XPS) to provide a cost-effective and sensitive nicotine detection material. LOD 0.149 µmol L−1 indicates the high accuracy in determining NIC levels [10]. Graphite carbon-nitride (g-C_3N_4) and multiwalled carbon nanotubes (CNTs) [11]; 2D hexagonal boron nitride nanosheets (BN) doped graphene film [12], were some of the other electrochemical sensors used to determine NIC.

4. ARTIFICIAL INTELLIGENCE AND MACHINE LEARNING IN BIOSENSORS

The integration of artificial intelligence in the medical and health sectors will mark a milestone in innovation, as data generated by machines and biosensors needs to be analyzed. This is because the data produced may not be suitable for traditional analysis methods due to its complexity. Not only complexity, but also noise, parameters, and other factors influence the use of artificial intelligence for solutions. Applications of artificial intelligence and machine learning algorithms in electrochemical data analysis have significantly improved the sensitivity and specificity of detection methods [13]. One of the breakthroughs in AI-integrated biosensing was Alphafold2 [14]. Meta AI entered the field by developing a model that predicts over 600 million proteins [15]. Powerful GPU clusters, such as NVIDIA-DGX, along with parallel processing technologies, have led to significant advancements in processing power and biosensor technologies [16]. Real-time monitoring of various biomarkers can be achieved using flexible electrochemical sensors combined with machine learning algorithms [17]. Machine learning is capable of learning from experience and adapting to new datasets. Using machine learning, AI systems gain the ability from experience, which enables the creation of algorithms that enable the computer to learn things from its own data and experiences [18]. Machine learning is categorized into supervised, unsupervised, and reinforcement learning (Fig.1)

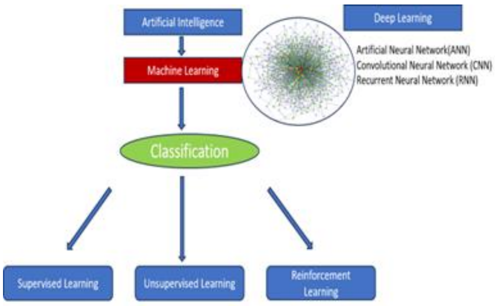


Fig 1: Artificial Intelligence, Machine Learning, and its categories.

Electrochemical diagnosis can be done using machine learning, but the data pre-processing is an essential step before training an ML model. The raw data should be easily understandable by the model to process. Normalization, Feature Extraction, Data Augmentation, etc, are the numerous pre-processing techniques [19]. AI-driven biosensors can be used to detect cancer-specific biomarkers in blood samples, and glucose level monitoring in diabetic individuals [20] [21]. beyond blood sugar monitoring, these biosensors are also employed to detect other biochemical markers in blood and urine, such as lactate, pyruvate, urea, and cholesterol biomarkers [22]. Moreover, they are also used in the detection of environmental and industrial pollutants like nitrate, nitrite, and sulfate [23].

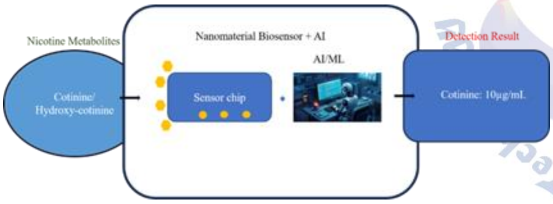


Fig 2: Schematic representation of Smart Biosensor Platforms for Nicotine Metabolite Detection.

ML can predict smoking-induced non-communicable diseases like lung cancer and postmenopausal osteopo rosis by identifying biomarkers and genetic profiles [24]. Researchers are using ML to understand psycho-genetic predispositions to tobacco addiction, providing personalized tools for quitting, monitoring unregulated vaping trades in social media by analysing behavioural p atterns in people who use tobacco [25] [26]. A major cause of lung cancer is prolonged exposure to tobacco, with an increasing risk with more years of smoking [27]. Many long-term smokers never develop lung cancer; m oreover, lung cancer also occurs in non-smokers, which is often diagnosed at the final stages, indicating the complex interaction of the body with tobacco. The significant gap mentioned that there is a need to identify risk pathways of smoking carcinogenesis for early detection of lung cancer and offering personalized treatment, especially for non-smokers [28]. Chen and Lin, 2020 investigated and reported that using machine learning, the impact of smoking factors on lung cancer can be identified, and also identified specific risk pathways associated with smoking-induced lung cancer [29].

5. PROPOSED SMART BIOSENSOR

5.1 Surface-enhanced Raman Spectroscopy (SERS)

Surface-enhanced Raman Spectroscopy (SERS) is a hig hly sensitive detection technique that provides structural and chemical information about the target molecule. Using silver or gold nanoparticles, SERS can detect nicot ine at nanomolar concentrations, making it an ideal analytical sensor for detecting NIC in complex biological samples like saliva and blood [30]. SERS is a powerful emerging technique that significantly enhances the detection limit of Raman scattering when the target analyte is adsorbed onto metallic nanostructures acting as antennas, amplifying the signal while preserving all v ibrational spectroscopy features [31]. Gold and silver nanoparticles are commonly used by the SERS community due to their cost-effectiveness and ease of manipulation, while offering a high enrichment factor. Gold nanoparticles are less affected by oxidation and are more uniform in size and shape. These advantages make them superior to silver nanoparticles, which are typically synthesized via chemical reactions [32] [33]. Compared to traditional and standard chromatography methods, SERS requires less extensive pre-processing, facilitating rapid field deployment. The size of the SERS sensor is one of the notable features, allowing it to be integrated with portable spectrometers and smartphone integration, which makes it suitable for IoT-enabled health services and point-of-care diagnostics [34].

5.2 IoT Hardware Architecture.

5.2.1 Sensing Module

The Surface-enhanced Raman Spectroscopy (SERS) interface collects data from the biological sample

5.2.2 Interface circuit

Electrochemical signals produced by the sensor are usually detected by amperometric and potentiometric measurements [35] [36]. An electric charge is produced due to the accumulation of charge molecules at the electrodes. Electrochemical sensors require a specific recognition of the target molecule to achieve detection specificity (e.g., antibodies, proteins, peptides). Electrochemical analysis techniques can utilize various functional units of a smartphone such as its power supply, wired data transmission interfaces like USB connectors, audio port-based connectors, and wireless peripherals like Bluetooth and Near Field Communication (NFC) to generate excitation signals and perform signal quantification [37] [38]. Electrochemical analysis techniques can also be integrated into smartphones via wired peripherals, such as audio connectors and Universal Serial Bus (USB) connectors [38]. Modes of electrochemical detection: the proposed system can perform electroanalytical techniques like 1. Cyclic voltammetry (CV), 2. Chronoamperometry, 3. Differential pulse voltammetry (DPV), 4. Square wave voltammetry (SWV) and 5. Potentiometry.

5.2.3 Microcontroller/Communications Board

Using the ESP32 Microcontroller, the voltage signal thus produced can be transferred to a smartphone via Blu etooth, Wi-Fi, or NFC. The signal thus produced in the ESP32 microcontroller, ESP32 Microcontroller, generates a sweep voltage signal, sent to the analog circuit, so the measurement can be read in the ESP32 [39] (Fig.3).

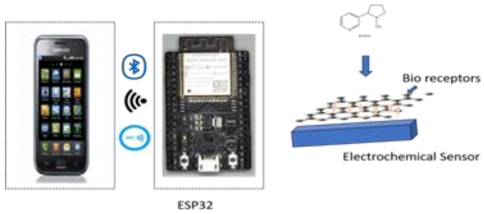


Fig 3: Schematic representation of the working of the ESP32 Microcontroller integrated with a smartphone for electrochemical sensing.

5.2.4 Smartphone Integration

The smartphone app receives the real-time processed data from the microcontroller through either Bluetooth, Wi-Fi, or NFC. That application detects nicotine levels, tracks trends, and, of course, correlates these with behavioral data such as smoking, medication, and geolocation for personalized health insights. The processed data can be uploaded and stored in the cloud securely for remote monitoring, telemedicine consultation, or research data aggregation, with user consent and encrypted channels [40].

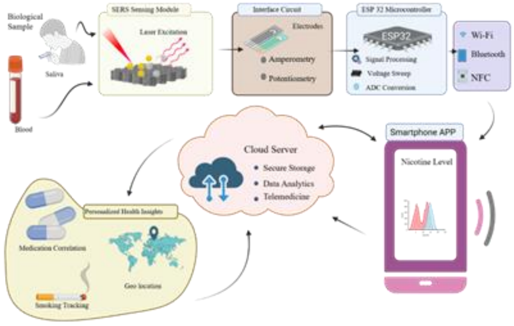


Fig 4: Schematic representation of the proposed detection model integrated with Artificial Intelligence and Machine Learning

6. DISCUSSION

The emergence of nanomaterial-based biosensors integrated with artificial intelligence (AI) and machine learning (ML) offers a transformative pathway for real-time detection of tobacco metabolites, particularly nicotine and its primary metabolite, cotinine. Traditional laboratory analytical techniques such as gas chromatography–mass spectrometry (GC–MS), high-performance liquid chromatography (HPLC), and enzyme-linked immunosorbent assays (ELISA) have been the gold standard for biomarker detection. However, they were limited by their high cost, bulky infrastructure, sophisticated techniques, long processing time, bulky infrastructure and need of trained personnel [3].These constraints make them unsuitable for continuous or point-of-care (POC) monitoring, especially in low-resource settings. The reviewed studies collectively suggest that electrochemical biosensors, particularly those employing nanostructured materials, can address these challenges. Electrochemical sensors using nanostructured electrodes such as nitrogen-doped graphene sheets [41], titanium dioxide–carbon paste composites [8], and Au nanostructure-modified TiO [10] have demonstrated exceptional sensitivity and low detection limits. These sensors exploit the high surface area-to-volume ratio, conductivity, and catalytic activity of nanomaterials, which enhances electron transfer k inetics and binding efficiency at the electrode interface [4]. Such attributes are critical when measuring trace metabolite concentrations in complex biological matrices like saliva, urine, and blood, where non-specific adsorption and matrix interference are common.

Additionally, the use of flexible substrates, paper-based platforms, and 3D-printed architecturess [42] [9] further improves the portability, cost-effectiveness, and field-deployability of these sensors, making them more suitable for consumer health monitoring. Despite these advances, electrochemical biosensors face challenges in data interpretation due to complex and often noisy electrochemical signals. This is where AI and ML integration provides a substantial advantage. ML algorithms can learn patterns from large datasets, filter noise, extract relevant features, and predict analyte concentrations with high accuracy [13]. Various models supervised (e.g., support vector machines, random forests), unsupervised (e.g., clustering), and reinforcement learning can be trained to recognize voltammetric, amperometric, or impedance signatures associated with nicotine metabolites [18]. Such approaches enable robust classification and quantifi cation even when raw signals are affected by user variability, electrode fouling, or environmental conditions. Moreover, the use of AI extends beyond analytical performance. As shown by Chen and Lin, 2020 ML can identify risk pathways linking smoking exposure to lung cancer, while Sinha et al, 2024 demonstrated that ML models can predict smoking-related diseases such as postmenopausal osteoporosis. Integrating these predictive health analytics into biosensing platforms could transform them from mere diagnostic devices to comprehensive preventive health systems. This aligns with broader digital health trends where biosensors are coupled with smartphone-based data collection, cloud storage, and personalized feedback, fostering user engagement and behavioral change. Smartphone integration also plays a pivotal role in the proposed biosensor architecture. Electrochemical signals generated during sensing can be powered and read using a smartphone's built-in fun ctional units power supply, USB/audio connectors, and wireless peripherals like Bluetooth and NFC [37] [38]. Microcontrollers like ESP32 can interface with the biosensor to transmit processed data to mobile applications in real time [39]. Such mobile health (mHealth) approaches leverage the ubiquity and computational power of smartphones, enabling decentralized diagnostics, telemedicine, and continuous monitoring outside clinical settings [40]. Importantly, the portability and low cost of such integrated systems make them suitable for large-scale epidemiological surveillance, especially in low- and middle-income countries where tobacco-related disease burdens are high. Surface-enhanced Raman spectroscopy (SERS) further enriches the toolbox for nicotine metabolite detection. By using metallic nanostructures like gold and silver nanoparticles, SERS achieves molecular fingerprinting with extremely low detection limits [30] [31]. Unlike conventional chromatographic methods, SERS requires minimal sample preparation, operates rapidly, and can be miniaturized for field deplo yment [34]. This makes it a strong candidate for multiplexed detection of multiple biomarkers from small sample volumes. However, standardizing SERS substra tes and ensuring reproducibility remain key challenges for clinical translation [32] [33]. Despite these promising technological advances, several translational challenges remain. First, the reproducibility of nanomaterial synthe sis is crucial, as batch-to-batch variation can affect sensor performance. Second, the stability and shelf life of biological recognition elements (aptamers, antibodies, molecularly imprinted polymers) must be optimized for real-world use. Third, regulatory and ethical considerations must be addressed for AI-driven health devices, particularly regarding data privacy, model bias, and clinical validation. These challenges underscore the need for interdisciplinary collaboration among material scientists, biomedical engineers, data scientists, and regulatory agencies. Nevertheless, the convergence of nanotechnology, electrochemical biosensing, and AI/ML represents a paradigm shift in tobacco metabolite monitoring. This approach could enable frequent, non-invasive, and user-friendly monitoring of nicotine exposure, empowering both clinicians and individuals to track smoking behavior, assess risk, and support cessa tion efforts. Furthermore, integrating behavioral and geolocation data via smartphone apps could open avenues for precision public health strategies, such as real-time exposure mapping and targeted interventions for high-risk populations [25] [26].

7. CONCLUSION

This review highlights the potential of next-generation biosensors that synergistically integrate n anomaterial-based transducers with AI/ML-driven data analytics for nicotine metabolite monitoring. Compared to conventional laboratory-based assays, these smart biosensors offer distinct advantages like miniaturization, portability, cost-effectiveness, real-time response, and seamless integration with digital health ecosystems. Electrochemical sensors using advanced nanomaterials such as graphene, carbon nanotubes, and metal nanoparticles have already demonstrated remarkable sensitivity and low detection limits, while SERS offers molecular-level specificity with rapid analysis times. Coupling these sensing platforms with AI/ML algorithms enables automated feature extraction, noise reduction, and accurate prediction of metabolite concentrations, thereby overcoming key limitations of traditional approaches. Importantly, smartphone-enabled biosensors bridge the gap between laboratory diagnostics and point-of-care applications. By leveraging smartphones' built-in power, communication interfaces, and computing capabilities, biosensors can become accessible and scalable tools for population-wide nicotine exposure monitoring. This has profound implications for smoking cessation programs, public health surveillance, and early disease risk prediction. Furthermore, AI models embedded within such platforms can continuously learn from user data, offering personalized feedback and behavioral insights, and even predicting downstream health outcomes such as lung cancer or osteoporosis. However, realizing this vision requires addressing several critical challenges. Standardizing nanomaterial fabrication methods, improving the long-term stability of biorecognition elements, and ensuring the robustness and interpretability of AI models are essential for regulatory approval and clinical adoption. Data security and user privacy must be safeguarded through encryption, anonymization, and transparent consent frameworks. Additionally, interdisciplinary research collaborations and public-private partnerships will be necessary to accelerate the translation of these technologies from proof-of-concept prototypes to widely deployed health tools. In conclusion, the fusion of nanotechnology, biosensing, AI/ML, and mobile health has the potential to revolutionize tobacco metabolite monitoring. Such integrated platforms could enable proactive, personalized, and preventive healthcare interventions, ultimately reducing the global burden of tobacco-related diseases. Future research should focus on developing standardized protocols, conducting large-scale clinical validations, and creating open-access datasets to train and benchmark AI models. With continued innovation and collaboration, smart biosensors for nicotine metabolites can evolve from experimental concepts into transformative tools for public health and precision medicine.

Conflict of interest statement

Authors declare that they do not have any conflict of interest.

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