Review Article

Recent Trends in Advanced Biosensors for Early Detection of Fungal Spoilage and Mycotoxication in Food of Animal Origin

Samah Eid[1], Elsayed S.E. Shabana[2], Amany N. Dapgh[3], Ahmed Shabaan[2], Ali Amer[3], Asmaa E. Gamal El-deen[3], Wagdy S.B.Youssef[3], Ashraf S. Hakim[4] and Hussien A. Abouelhag[4]

[1]Reference Laboratory for Veterinary Quality Control on Poultry Production, Animal Health Research Institute, Agriculture Research Center (ARC), Giza, Egypt.

[2]Department of Food Hygiene, Animal Health Research Institute, Agriculture Research Center (ARC), Dokki, Giza, Egypt.

[3]Bacteriology department, Animal Health Research Institute, Agriculture Research Center (ARC), Dokki, Giza, Egypt.

[4]Department of Microbiology and Immunology, National Research Centre, 33 Bohouth St., Dokki, Cairo, Egypt.

Corresponding author: Amany N. Dapgh **E-mail:** amany.nabil.dapgh@gmail.com

Received: 29-07-2025, **Accepted:** 16-08-2025. **Published online:** 23-08-2025
DOI: https://doi.org/10.33687/ricosbiol.03.08.68

Abstract

The public health concern induced by mycotoxins contamination has globally retained a prominent deal of interest. Mycotoxins are secondarily synthetized and can accumulate in host organs so institute adverse effects on humans and livestock, resulting in grave health threats and often produced by certain filamentous fungi broadly found in foodstuffs. Enhancing early trace recognition and control from the root is a more coveted approach than the disposal way to assert food safety. Biosensors are ready to interference from various components in intricate food matrices when recognizing trace mycotoxins. This article focuses on advanced approaches especially incorporation of biosensors for detection of mycotoxins in food matrices of animal origin as well as progressing of sensing detection for food safety assurance.

Key words: Mycotoxins, milk, meat, food safety, biosensors.

Introduction

Mycotoxins are toxic secondary metabolites formed as secondary metabolites by different filamentous fungal species and produced under particular circumstances (Pandey et al., 2023). According to the previous reports, there are more than 500 mycotoxins that have been recognized as toxigenic and harmful to plant, animal, and human health and so far, could be differentiated to groups following their own toxic impacts. The most prevalent

established types involve Aflatoxins (AFTs), Citrinin (CT), Fumonisins (FUMs), Ochratoxins (OTs), Patulin (PAT), Trichothecenes (TCTs), and Zearalenone (ZEN) (Haque *et al*., 2020).

Mycotoxins compress a set of structurally assorted low molecular weight chemical compounds, commonly less than 1000 Da produced when temperature within the range of 25 ± 5 °C. Additionally, water activity is also a substantial factor affecting mycotoxin synthesis, which informs the quantity of water attainable for microbial and chemical paths within a substance like food (Janik *et al*., 2020).

Mycotoxins are sorely poisonous and their consumption may result in acute or chronic health issues. Mycotoxicosis, has accompanied with signs may vary relying on the type of mycotoxin and may comprise the persuading: cytotoxicity, hepatotoxicity, nephrotoxicity, neurotoxicity, teratogenicity, and carcinogenicity (Pandey *et al*., 2023).

Since the premier discovery of mycotoxins, many analytical techniques have been scouted and employed for estimating their existence in food and feed. Chromatographic methods have often been utilized, due to their versatility, these comprise thin-layer chromatography (TLC) and high-performance liquid chromatography (HPLC), in coupling with a scope of detectors such as diode array, fluorescence and UV, as well as, gas chromatography–tandem mass spectrometry (GC-MS/MS) and liquid chromatography–tandem mass spectrometry (LC-MS/MS). Moreover, it is also noting the worthy role of antibody-based immunoassays in mycotoxin recognition (Yang *et al*., 2020).

Essentially, a biosensor is an advanced analytical tool that combines biological sensing components, like antibodies, enzymes, organelles even whole cells or tissues with a transducer that known as bioreceptor. The bioreceptor segment which also termed as 'the detector element' has a selective site able to define the target and converts the biological interaction into a detectable signal, such as an electrical, optical, or thermal output. This measurable signal is directly proportional to the concentration of the specific analyte or group of analytes of concern. The type of transducer used relies on the biosensor's specific outlay while the type of biosensor is defined by transducer mechanism (Gaudin, 2017).

Methodology

1. *Running Approaches in Mycotoxins Recognition*

1.1. Sampling

Following to present knowledge, the readiness of a specimen for identification requires the achievement of procedures involving sampling, grinding, mixing, extraction, and purification. The contamination of natural, solid products with mycotoxins is non-homogeneous and may display random allocation, possibly resulting in false -ve data and the failure to recognize presenting threats when sampling from improper areas is carried out (Zhang *et al*., 2018).

1.2. Sample Readiness: Extraction and Purification

After sampling, the specimen should be ground and mixed to permit and speed up the chemical reaction processes. Relied on the previous literature, the homogenized particles' final size should be 500 µm approximately (Nakhjavan *et al*., 2020). There are abundant various methods for extracting mycotoxin, and the proper choice of the pretreatment approach is necessary because of the different consistencies of food products. Relied on the traditional methods, the most prevalent selected are Solid–Liquid Extraction (SLE), Solid-Phase Extraction (SPE), and QuEChERS.

SLE is one of the most prevalent tehniques used to extract mycotoxins from various foodstuffs. SLE is easy to conduct and does not necessitate large financial budgets and any particular instruments. However, to obtain precise and perfect results, the solvent must be carefully chosen (Bian *et al*., 2023). ii. Solid-Phase Extraction (SPE) is an efficient technique used to extract mycotoxins. The liquid sample that comprises the analytes of interest is passed via the unique cartridge that includes high-affinity adsorbent particles. SPE is superior to other conventional methods, as it minimizes solvent usage, efficient concentration, and promoted recovery rates (Badawy *et al*., 2022). iii. QuEChERS, this doubled technique name is interestingly coming from the persuading terms: Quick, Easy, Cheap, Effective, Rugged, and Safe (Pereira *et al*., 2015). Like in the formerly described technique, the key of the good optimization of QuEChERS is a good choice of sorbents (Badawy *et al*., 2022).

2. *Techniques Used for Mycotoxin Recognition*

2.1. Thin-Layer Chromatography (TLC) is a type of liquid chromatography that was broadly employed between in the last twenty years in the past century and still in use today because of its low analysis cost. TLC employs a stationary phase typically formed of silica, cellulose, or alumina, pasted to an inert material such as plastic or glass that retains the analyte in place through separation. Meanwhile, the mobile phase, often including acetonitrile, methanol, and water mixtures, transmits the specimen across the stationary phase (Meyers and Meyers, 2008). The visualization of TLC can be classified into three prime categories: destructive (chemical compounds, ninhydrin, bromocresol green, and p-methoxy benzaldehyde), semi-destructive (iodine staining), and non-destructive (UV staining) (Ventura *et al*., 2005).

2.2. Liquid chromatography (LC) is one of sensitive, more specific, and automated techniques developed to overcome the disadvantages of TLC. LC permits the simultaneous recognition of many mycotoxins, however of their biological activity and chemical composition. LC is more efficient in identifying mycotoxins but it necessitates remarkably greater financial budget, involving the purchase of convenient equipment (Yang *et al*., 2020).

2.3. High-performance liquid chromatography (HPLC) is a gold-standard technique in the assessment of mycotoxin contamination in different foodstuffs. The guidelines for their detection greatly follow similar techniques, using fluorescence detectors, UV–visible or even mass spectrometry to enhance the sensitivity and effectiveness (Turner *et al*., 2009).

2.4. Gas Chromatography (GC) is a chromatography type of which the mobile phase is gas. GC is less used for the recognition and quantification of mycotoxins in food specimens so a commercial protocol for GC not obtained, especially with the presence of faster and cheaper methods like HPLC (Rodríguez-Carrasco *et al*., 2014).

2.5. Enzyme-linked immunosorbent assay (ELISA) is considered one of the most prevalent employed antibody-based immunoassays for mycotoxin determination. ELISA exposes the simple, rapid, reliable, and simultaneous analysis of numerous specimens. ELISA kits are usually relied on a competitive assay format and characterized by its high specificity, portability and fast execution time. ELISA kits are limited for single use, which may elevate the cost of conducting a screening assay of abundant mycotoxins (Maggira *et al*., 2022).

2.6. Lateral flow immunoassay (LFA) is a low-cost, simple, paper-based antibody-based immunoassay test for the rapid detection and quantification of different analytes as mycotoxins, aflatoxin M1 in milk (Singh *et al*., 2022).

3. *Mycotoxin Detection Techniques*

3.1. Fluorescence sensors which detect target analytes depending on the absorption and subsequent re-emission of photons by excited atoms or molecules either via their inherent fluorescence or through conjugation with a fluorophore (Lu *et al*., 2016). Fluorescent sensors are featured by their sensitivity, affordability, and rapid response time so offer the accurate identification and quantitative measurement of food contaminants as well as toxin recognition in milk (Naz *et al*., 2025).

3.2. Electrochemiluminescence, (ECL), is a type of luminescence sensor output during electrochemical reactions in solutions, the prime advantage of this technique is that there is no necessity for an excitation light source. ECL reactions are precisely controlled by electrical potential and outright in processes. These features make ECL technology an excellent tool for recognizing tiny amounts of toxins in food and the environment, as well as for diagnosing illnesses (Lv *et al*., 2023). Relying on the kinds of sensing components utilized, ECL biosensors can be classified into 3 groups, based on antibodies, aptamers, as well as molecular imprinting polymers (MIPs).

3.2.1. Immunosensors, employing antibodies as detection elements, display heightened specificity and sensitivity arisen from the unique merits of the binding between antigens and antibodies carried on nanoparticles (Li *et al*., 2021). These nanoparticles gave anchoring sites for immobilizing another antibody and served as efficient carriers (Lv *et al*., 2023). Immunosensors were applied for the precise detection of Aflatoxin M1 (AFM1) in milk and dairy products (Angelopoulou *et al*., 2023) as well as T-2 toxin in swine meat (Wang *et al*., 2018). Otherwise, there are existed challenges and limitations to be addressed in the utilization of immunosensors. Antibody bioactivity is sensitive to environmental circumstances, and this led to notable possibility of cross-reactions between antibodies and other biomolecules. This concern results in inaccurate data, involving both false positives and

false negatives during real specimen analysis. Therefore, it is fundamentally, that the accuracy and reliability of recognition are further modulated and developed in future studies, contribute to reliable and precise mycotoxin detection (Szelenberger *et al*., 2024).

3.2.2. Aptamer-Based Biosensor are short, single-stranded RNA or DNA (ssRNA or ssDNA) molecules that can selectively bind to a particular target, involving proteins, toxins, small molecules, and even live cells. In converse to antibodies, aptamers are not susceptible to temperature and are chemically stable, therefore, it is a promising ECL approach particularly for its prospect of coupling nanoparticles featuring unique physical and chemical merits to the terminal of nucleic acids (Jia *et al*., 2022). Updated various DNA hybridization techniques have been utilized in mycotoxin detection, Aflatoxin B1, Aflatoxin M1in milk and fumonisin B_1 in meat (Ahmadi *et al*., 2022, Ramezani *et al*., 2022 and Sun *et al*., 2023).

3.2.3. Molecular Imprinted polymers (MIPs) have acquired fundamental attention for their peculiar advantages, comprising exceptional selectivity, rapidity, reusability, and simplicity. Molecular imprint plays a pivotal role in these detection processes by achieving specific cavities that mimic the structure and shape of target molecules. These unique properties have resulted in the employment of MIP approaches for the efficient label-free recognition of mycotoxins, aflatoxins (Díaz-Bao *et al*., 2016) and zearalenone (Yugender Goud *et al*., 2019). In spite of some drawbacks, average sensitivity, a restricted ability to detect macromolecular targets, and complex preparation steps, MIPs have continued progresses and enhanced the abilities to meet analytical requirements across industries (Szelenberger *et al*., 2024).

4. *Incorporated Biosensor Implementations in Food Safety Management*

Food safety links to the assertion that food, when readiness and consumed as proposed, does not induce harm upon the consumer (Sorbo *et al*., 2022). Legislation intending food safety in developing countries is broadly not established, constituting a concern of health protection. The overall principle is to utilize an incorporated approach, involving all sides of the food chain, from farm to fork. Attachment to food safety requirements the systematic management of food hygiene and standards, asserting that the food products supplied are considered safe for consumption so it is necessary to establish a scientific basis for risk management (Lizakowski, 2019).

Naturally sourced food products often load microbiological risks. Even with significant progress in food safety, microorganisms remain the biggest threat to what we consume. To address this, microbiological criteria provide guidelines for the acceptability of food products and their production processes (Sosnowski and Osek, 2021). Preventive measures are substantial for food safety as Good Hygiene and Manufacturing Practices (GHP, GMP), Hazard Analysis Critical Control Points (HACCP) principles and Food Contact Material (FCM) Migration Testing (Eid *et al*., 2025).

Animal products such as meat, milk, eggs, or offal can be contaminated via the animals' diet. This highlights the need for a comprehensive approach to monitoring and controlling the existence of mycotoxins (El-Sayed *et al*., 2020). The use of biosensors in meat processing plants, encompassing slaughterhouses and dairies, can play a pivotal role in early recognition and prevention. By monitoring the existence of mycotoxins, these biosensors can aid and assert that meat and dairy products couple stringent safety standards prior to reaching consumers.

Conclusion

Biosensors are constantly improving and being integrated into food safety systems, showing that this is a rapidly changing field with huge potential to make our food supply safer and better. As technology, connectivity, and teamwork across different disciplines advance, we'll see even more uses for biosensors in keeping our food safe.

References

Ahmadi SF, Hojjatoleslamy M, Kiani H and Molavi H. (2022) Monitoring of Aflatoxin M1 in milk using a novel electrochemicalaptasensorbased on reduced graphene oxide and gold nanoparticles. Food Chem. 30,373(Pt A):131321.

Angelopoulou M, Kourti D, Misiakos K, Economou A, Petrou Pand Kakabakos S. (2023). Mach-Zehnder Interferometric Immunosensor for Detection of Aflatoxin M1 in Milk, Chocolate Milk, and Yogurt. Biosensors (Basel). 30,13(6),592.

Badawy M.E.I., El-Nouby M.A.M., Kimani P.K., Lim L.W. and Rabea E.I. (2022). A review of the modern principles and applications of solid-phase extraction techniques in chromatographic analysis. Anal. Sci.,38,1457–1487.

Bian Y., Zhang Y., Zhou Y., Wei B. and Feng X. (2023). Recent Insights into Sample Pretreatment Methods for Mycotoxins in Different Food Matrices: A Critical Review on Novel Materials. Toxins, 15,215.

Díaz-Bao M, Regal P, Barreiro R, Fente CA. and Cepeda A. (2016). A facile method for the fabrication of magnetic molecularly imprinted stir-bars: A practical example with aflatoxins in baby foods. J Chromatogr A., 1471,51-59.

Eid S., Gehad F.A. Fath Elbab , Amany N. Dapgh, Hakim A. S., Wagdy S.B. Youssef and Hussien A. Abouelhag. (2025). Food Contact Material (FCM) Migration Testing: Novel Assay for Ensuring Food Safety. Ricos Biology Journal, Vol. 3 (7) 12-16.

El-Sayed R., Jebur A., Kang W., El-Esawi M. and El-Demerdash F. (2020). An overview on the major mycotoxins in food products: Characteristics, toxicity, and analysis. J. Future Foods, 2,91–102.

Gaudin V. (2017). Advances in biosensor development for the screening of antibiotic residues in food products of animal origin—A comprehensive review. Biosens. Bioelectron, 90,363–377.

Yang Y., Li G., Wu D., Liu J., Li X., Luo P., Hu N., Wang H. and Wu Y. (2020). Recent advances on toxicity and determination methods of mycotoxins in foodstuffs. Trends Food Sci. Technol, 96,233–252.

Haque M.A., Wang Y., Shen Z., Li X., Saleemi M.K. and He C. (2020). Mycotoxin contamination and control strategy in human, domestic animal and poultry: A review. Microb. Pathog., 142,104095.

Janik E., Niemcewicz M., Ceremuga M., Stela M., Saluk-Bijak J., Siadkowski A. and Bijak M. (2020). Molecular Aspects of Mycotoxins—A Serious Problem for Human Health. Int. J. Mol. Sci., 21,8187.

Jia M., Jia B., Liao X., Shi L., Zhang Z., Liu M., Zhou L., Li D. and Kong W. (2022). A CdSe@CdS quantum dots based electrochemiluminescence aptasensor for sensitive detection of ochratoxin A. Chemosphere, 287,131994.

Li M., Yue Q., Fang J., Wang C., Cao W. and Wei Q. (2022). Au modified spindle-shaped cerium phosphate as an efficient co-reaction accelerator to amplify electrochemiluminescence signal of carbon quantum dots for ultrasensitive analysis of aflatoxin B1. Electrochim. Acta., 407,139912.

Lizakowski P K.A. (2019). Legal regulations and administration of food safety. World Sci. News., 122,133–144.

Lu S., Li G., Lv Z., Qiu N., Kong W., Gong P., Chen G., Xia L., Guo X., You J. and *et al.* (2016). Facile and ultrasensitive fluorescence sensor platform for tumor invasive biomarker beta-glucuronidase detection and inhibitor evaluation with carbon quantum dots based on inner-filter effect. Biosens. Bioelectron, 85,358–362.

Lv X., Tan F., Miao T., Cui B., Zhang J., Fang Y. and Shen Y. (2023). In situ generated PtNPs to enhance electrochemiluminescence of multifunctional nanoreactor COP T(4)VTP(6) for AFB(1) detection. Food Chem., 399,134002.

Maggira M, Sakaridis I, Ioannidou M and Samouris G. (2022). Comparative Evaluation of Three Commercial Elisa Kits Used for the Detection of Aflatoxins B1, B2, G1, and G2 in Feedstuffs and Comparison with an HPLC Method. Vet Sci., 9(3),104.

Meyers C.L.F. and Meyers D.J. (2008). Thin-Layer Chromatography. Curr. Protoc. Nucleic Acid. Chem., 34, A.3D.1–A.3D.13.

Nakhjavan B., Ahmed N.S. and Khosravifard M. (2020). Development of an improved method of sample extraction and quantitation of multi-mycotoxin in feed by LC-MS/MS. Toxins, 12,462.

Naz I, Alanazi SJF, Hayat A and Jubeen F. (2025). Covalent organic framework-based aptananozyme (COF@NH_2 apt-AFM_1): A novel platform for colorimetric and fluorescent aptasensing of AFM_1 in milk. Food Chem., 484,144478.

Pandey A.K., Samota M.K., Kumar A., Silva A.S. and Dubey N.K. (2023). Fungal mycotoxins in food commodities: Present status and future concerns. Front. Sustain. Food Syst., 7,1162595.

Pereira V., Fernandes J. and Cunha S. (2015). Comparative assessment of three cleanup procedures after QuEChERS extraction for determination of trichothecenes (type A and type B) in processed cereal-based baby foods by GC–MS. Food Chem., 182,143–149.

Ramezani M, Jalalian SH, Taghdisi SM, Abnous K and Alibolandi M. (2022). Optical and Electrochemical Aptasensors for Sensitive Detection of Aflatoxin B_1 and Aflatoxin M_1 in Blood Serum, Grape Juice, and Milk Samples. Methods Mol Biol., 2393,417-436.

Rodríguez-Carrasco Y., Moltó J.C., Berrada H. and Mañes J. (2014). A survey of trichothecenes, zearalenone and patulin in milled grain-based products using GC–MS/MS. Food Chem.,146,212–219.

Singh H, Singh S, Bhardwaj SK, Kaur G, Khatri M, Deep A and Bhardwaj N. (2022). Development of carbon quantum dot-based lateral flow immunoassay for sensitive detection of aflatoxin M1 in milk. Food Chem., 1 393,133374.

Sorbo A., Pucci E., Nobili C., Taglieri I., Passeri D. and Zoani C. (2022). Food Safety Assessment: Overview of Metrological Issues and Regulatory Aspects in the European Union. Separations, 9,53.

Sosnowski M. and Osek J. (2021). Microbiological Safety of Food of Animal Origin from Organic Farms. J. Vet. Res., 65,87–92.

Sun Y, Lv Y, Zhang Y and Wang Z. (2023). A stimuli-responsive colorimetric aptasensor based on the DNA hydrogel-coated MOF for fumonisin B_1 determination in food samples. Food Chem., 403,134242.

Szelenberger R, Cichoń N, Zajaczkowski W and Bijak M. (2024). Application of Biosensors for the Detection of Mycotoxins for the Improvement of Food Safety. Toxins (Basel), 27 16(6),249.

Turner N.W., Subrahmanyam S. and Piletsky S.A. (2009). Analytical methods for determination of mycotoxins: A review. Anal. Chim. Acta, 632,168–180.

Ventura M, Anaya I, Broto-Puig F, Agut M and Comellas L. (2005). Two-dimensional thin-layer chromatographic method for the analysis of ochratoxin A in green coffee. J Food Prot., 68(9),1920-2.

Wang Y, Zhang L, Peng D, Xie S, Chen D, Pan Y, Tao Y and Yuan Z. (2018). Construction of Electrochemical Immunosensor Based on Gold-Nanoparticles/Carbon Nanotubes/Chitosan for Sensitive Determination of T-2 Toxin in Feed and Swine Meat. Int J Mol Sci., 19(12),3895.

Yugender Goud K., Sunil Kumar V., Hayat A., Vengatajalabathy Gobi K., Song H., Kim K.-H. and Marty J.L. (2019). A highly sensitive electrochemical immunosensor for zearalenone using screen-printed disposable electrodes. J. Electroanal. Chem., 832,336–342.

Zhang L., Dou X.-W., Zhang C., Logrieco A.F. and Yang M.-H. (2018). A review of current methods for analysis of mycotoxins in herbal medicines. Toxins, 10,65.