11 Onsite Quality Controls for Food Safety Based on Miniaturized Biosensing

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Abstract

Health and wellness are linked to the food we regularly consume. Although the emergence of advanced technologies, such as intelligent packaging, safety, and transportation in temperature-controlled containers, has greatly improved the quality of food, certain microbial invasions and deliberate adulterations are unavoidable when it involves long transportation and extensive commercialization. These activities eventually make the food unhealthy to consume. Onsite quality control (QC) tends to check such food items not only to control the food spoilage in commercialization but also to protect the consumers from the consumption of unhealthy food. In this context, advances in miniaturized sensing devices have paved numerous possibilities to monitor food quality in an onsite context. This chapter discusses the existing ways of ensuring quality assurances

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for food safety and their associated challenges. Thereby, common indicators of quality and spoilage in different types of food items have comprehensively been described as control of the different types of food. Thereafter, the miniaturized biosensor-based devices for food quality assurance have been described, where a brief discussion on the development processes, analytical performances, and commercial potentials are discussed with various examples and reported potential products for food quality assurance and safety.

Keywords

Food quality · Food safety · Miniaturized devices · Food sensors · Onsite quality control

11.1 Introduction to Packaged Food and the Need for Onsite Quality Controls

Over the last five decades, the food industries have grown exponentially to meet the needs of the vast majority of the population. Since then, packaged, ready-to-eat food has come into our habits and lifestyle. Unfortunately, many of these ready-to-eat processed foods are compromised with nutritional values and have preservatives in them, which are critical not only for health concerns but also for socio-economic impacts (Lozano et al. 2019). While the global demand for healthy food has been fulfilled by various farms and food processing companies, food safety is still posed as one of the biggest concerns. To ensure food safety, the governing bodies and policymakers have enforced strict regulation at various steps of production and commercialization processes including farm produce, food processing, storage, transportation, and retailing. However, the frequent occurrence of food-borne pathogenic outbreaks and mass food poisoning proves the hidden nature and threats to food safety (Mahato and Chandra 2019). Several such instances have been reported across the world in the recent past. For example, the *Escherichia coli* outbreak in Germany in 2011 and the mass sickness due to contaminated infant formula in China in 2008. Such occurrences not only cause casualties but also severely impact the economic condition, which may eventually result in a crisis (Lim and Ahmed 2016).

To avoid such disasters and improve the food habits, food technologists, scientists, and industries have been collaborating to address the issues and shortcomings by introducing a variety of detection strategies and techniques for fast, responsive, reliable, and inexpensive tests for checking the food quality and contaminating/spoiling agents (Ibrišimović et al. 2015; Lozano et al. 2019). For quality evaluations at the food safety assessment center and QCs, several instruments and techniques are commonly being employed, which include chromatography, spectrophotometry, and immunoassays-based assessments. Although these techniques offer highly sensitive and reliable determinations, several limitations viz. requirement of highly trained personnel, dedicated laboratory spaces, etc. are also associated with their application in onsite settings. Also, the usage of such high-end instruments adds the analysis costs to the commodities and thus greatly impacts the commercial values, especially in larger volumes (Mahato et al. 2017, 2018e).

In this context, the miniaturized tools capable of rapid and equally sensitive detections are of utmost need to fulfill onsite examinations of the food items. Among various such miniaturized detection devices, the biosensors-based detectors have found great attention for the detection of food quality and spoilage biomarkers to incorporate to realize onsite and efficient QCs. The biosensor-based modules detect the quality and safety biomarkers of food items by targeting the molecules, pathogenic determinants, or any chemical markers that are either responsible or arose due to the spoilage/contamination of the food. Such detections/monitoring have been done by the sensor probe, composed of the bio-receptors, which are governed by various interactions viz. receptor/ligand, antigen/antibody, enzyme/substrate, nucleic acid hybridization, and chemical interactions (Kumar et al. 2020a; Weston et al. 2021). The following section describes such processes of biosensor developments, their concerns for designing onsite food quality and safety assessment.

11.2 Food Biosensors: Design and Development

The biosensor is an analytical device composed of receptors and the transducer surface that generates the quantifiable signal upon interaction with the analyte molecule (Kashish et al. 2017; Mahato et al. 2021). The commonly used receptors for the detection of biomarkers are antibody, cell, and enzyme, which are coupled with various kinds of transducer surfaces viz. electrochemical, optical, and piezoelectrical (Mahato et al. 2016a; Prasad et al. 2016b). The transducer converts the bio-interactions of the food quality/spoilage markers with the sensor probe and produces quantifiable signals based on analyte concentrations. The choice of transducer surfaces is highly influenced by the nature of the analyte and the QC test that are to be made. For example, the qualitative estimation of the food spoilage markers is commonly done by the optical transducer, which mainly offers yes/no based detection. However, in case of quantification of the contents (that is, low sugar, high sugar, and percentage of alcohols) are mainly estimated by the sensitive electrochemical-based transducers (Mahato et al. 2016b, 2019). Estimating the nutrient levels in processed food requires high-frequency estimations and are commonly offered by electrochemical-based detections, owing to their reusable and inexpensive nature (Mahato et al. 2020a).

In the conventional QC centers, detections are mainly performed in a randomized manner, where the arbitrary sample is collected from a batch and checked in the centralized laboratory. These tests are mostly time-consuming, and resource-intensive, which has led to the development of minimalistic approaches for lessening the burden. This huge demand for onsite QCs has paved the way for exponential increment of biosensor-based research for quality assurance at onsite settings. This section describes various components of biosensors and detection techniques focusing on the types of markers for ensuring food quality and safety from the markers originated from the food items and the markers from environmental exposure (viz. bacteria, fungus, and other associated indicators), which play a key role in food spoilage.

11.2.1 Components of Biosensors

The major components of a biosensor include the bio-receptors, transducer, and processor (Mahato et al. 2018f). Bioreceptors are the biological molecules and are primarily interacting molecules (Purohit et al. 2019a). These interactions are broadly classified under two categories: biocatalytic and bio-affinity. The biocatalytic interactions generally use enzymes, whole cells, or tissue slices, which generate the electroactive species or the bi-product in presence of the analyte (Prasad et al. 2016b). Another type of bio-receptor is mainly for affinity-based detection, which follows the receptor-ligand conjugation strategies (Kumar et al. 2019b). These comprised the antibody, aptamer, affimer, and affibody-based strategies, which are highly specific receptors (Mahato 2019). These are highly specific and coupled with the traducer surface majorly in the electrochemically active transducer (Mahato et al. 2021). The impedance-based electrochemical technique is commonly used for such affinity-based interactions. This technique shows higher sensitivity, however, are more prone to the interferences, due to the presence interfering agents co-exist in testing samples, and their time requirement for the affinity binding limits from their usage in the rapid onsite detection of food quality or spoiling markers. In some cases, these are used where there is no chance of generating the electroactive species for quantifications (such as, estimation of toxins and bacteria). In recent times, the biosensors-based strategies for checking the marker molecules have been greatly recognized, and are constantly being improvised in terms of their analytical performance (Mahato et al. 2018b, f; Kumar et al. 2019c, 2020b; Mahato 2019; Purohit et al. 2020). Figure 11.1 depicts various components of the biosensors and the classification based on the bio-receptors and the transducer. The formal definition of biosensors is coined as the device that uses specific biochemical reactions mediated by isolated enzymes, immune systems, tissues, organelles, or whole cells to detect chemical compounds usually by electrical, thermal, or optical signals (Mahato et al. 2018a). The performances of biosensors have been evaluated in various parameters viz. selectivity, sensitivity, reusability, dynamic ranges, detection limit, etc.

To enhance the analytical performances, several transducer materials have been employed. Among all, nanomaterial-based transducers have widely been accepted due to their extremely powerful electron transfer capability, which eventually allows better sensitivity (Baranwal et al. 2016; Prasad et al. 2016a; Mahato et al. 2018c, 2019). In some cases, the adoptions of nanomaterials in biosensing mechanism have leveraged enhanced sensor performances in miniaturization for the detection in onsite settings with comparable performance to the gold standard techniques (Purohit et al. 2020b). In the recent past, various nanomaterials have been used for

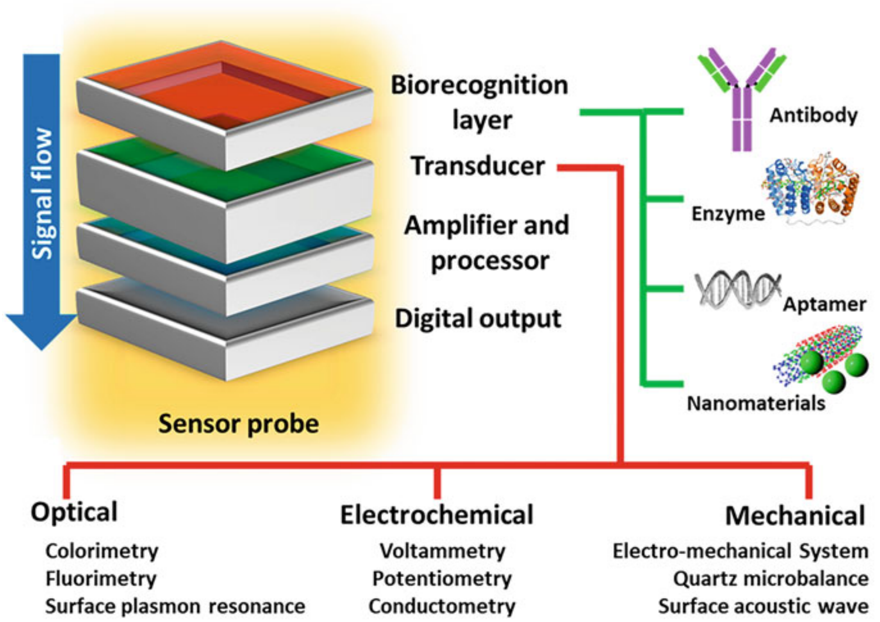

Fig. 11.1 Anatomy and the classification of the biosensors: (Left) components of the biosensor. (Right) different types of the bio-recognition layers, and (bottom) different types of biosensors based on the transducer surface

developing biosensing matrices, which include metallic nanoparticles, carbon-based nanomaterials, polymeric nanomaterials, quantum dotes, nanocomposites, etc. (Purohit et al. 2019b; Mahato et al. 2020d). Due to their size-dependent optoelectronic properties and nano-catalytic activity, these nanomaterials can be incorporated as recognition elements (Kumar et al. 2019a). The coupling of specific bio-receptors to the nanoparticle-based transducer has also been employed to introduce the selective detection of the analytes, where the covalent coupling/immobilization is preferred for better stability and reproducibility (Mahato and Chandra 2019). To fabricate the biosensors for onsite detection, various platforms have been used, among which lateral flow assay, dipsticks, electrochemical chip-based modules, and microprocessor-integrated wearable modules have found great attention for the biomedical detections and onsite- QCs (Teymourian et al. 2021; Mahato and Wang 2021). So far, different types of techniques have been used for developing biosensors-based modules for food quality assurances and their safety. Among these, optical and electrochemical-based strategies are widely accepted in food safety and quality detection due to their ease of usage and cost-effectiveness. This section describes various techniques that have been employed for detecting the biomarkers of food quality and spoilage.

11.2.1.1 Optical Biosensing Techniques

In optical biosensors, the optical properties of the transducers are commonly exploited to detect/quantify the analytes, which exhibit perceivable optical signals upon the interaction of the target analyte with the sensor probe. Due to their simple and non-destructive operation, these are the first choice for quality estimations and screening. Optical biosensing techniques are most widely used for the qualitative testing of food quality and spoilage. There are various formats adopted for the optical-based determination, which rely on color change, fluorescence change, and surface plasmon alteration (Mahato et al. 2020b), using an interferometer, resonators, garters, refractometers, etc. (Estrela et al. 2016; Kumar et al. 2020a). The nanomaterial-based colorimetric detection involves the recognition of the analyte, which upon the interaction changes the color of the working surface/area and is correlated to the concentrations (Kumar et al. 2015). The color change could be perceivable by the naked eye and aided eye for qualitative and detailed quantification, respectively. Since the colorimetric sensors offer easy handling, this format has widely been adopted among the other optical-based sensing formats (Estrela et al. 2016). The operation simplicity offers the best suitability in on-site QCs and collection centers where the batches of food items are being produced, packaged, or processed.

11.2.1.2 Electrochemical Biosensing Techniques

Although the preliminary screening saves the food items from mass spoilage, it does not confirm the prolonged shelf-life, as these are prone to get invaded by microbes during storage and transportation. Thus, the periodic evolution of the food items is necessary to achieve a prolonged shelf-life. The major limitation of most of the optical-based detections is for single-use, which limits its periodic usage to fulfill the constant/periodic tracking of the biomarkers. However, multiple sensors can be used for fulfilling the purposes, but that would certainly add more cost. Hence, electrochemical detections are being adopted for mitigating such challenges, which not only offer better sensitivity but also are capable of delivering stable detection. These are advantageous because of their easy integration to the analyzer modules, incorporating the onsite quantifiable detection with ease. The commonly used electrochemical-based techniques are voltammetry, amperometry, and impedimetric sensing as most of the bio-analytical reaction either produces the conductance or the impedance. In the format where the bioanalytical reaction produces electroactive species, voltammetry of the amperometry/conductometry is being used for the detection, while the bioanalytical reaction does not produce electroactive species, impedimetric-based approaches are commonly used, such as receptor–ligand interactions. For instance, a toxin found in milk “aflatoxin M1” has been detected employing the immune-complexation process followed by the impedance recording, which changes when the receptor and ligand bind at the sensor probe surface.

11.2.2 Indicators of Food Quality

Quality is a crucial attribute for any food processing industry, which determines the company's growth in this competitive market. The food industries can broadly be categorized under "beverages," "dairy," "meat," and others. The beverages are recreational food items and are mainly alcoholic and non-alcoholic sugar-sweetened beverages. For the quality improvement of the alcoholic beverages, mainly, ethanol, lactic acid, malic acid, polyphenols, and glycerol contents are detected, while in the sugar-sweetened beverages glucose, fructose, aspartame, and ascorbic acid have been commonly estimated for evaluating the quality. The dairy industries have a wide range of products and thus have various indicators for improving the quality of the specialized variant of a product. However, in all cases, lactose has been the major indicator of dairy food quality. Similar to beverage industries, another major sector is covered by the meat industries, where the meats (viz. seafood, fish, chicken, beef, pork, etc.) of various kinds are processed for maintaining the quality for a longer time and are sold in the market. The major challenge in quality assurance for such processed meats are their improper processing, additives and preservatives. As amines and nitrates are commonly employed for the preservatives and antioxidants at the meat curing process, the nitrate content provokes in situ nitrite formation in meat, which has reported of having carcinogenic properties. Thus, the estimation of amine and nitrate in the meat is essential for its quality improvement. Apart from this, in seafood meats, the number of heavy metals is more prominent, thus the estimations of toxic heavy metal ions are much important to ensure the quality of the food. Bakery industries are another popular food sector that uses grains, flour, and additives as common raw materials in preparing the recipes, which has prolonged the shelf life. Due to the involvement of high-temperature heating, the chances of the formation of toxicants (acrylamide which forms in the presence of aliphatic amide and asparagine) are very high, thus acrylamide detection is used as a major biomarker in bakery industries.

11.2.3 Indicators of Food Security

To sustain the food for a longer time, preservatives and antibiotics are commonly added during the processing and packaging. While there are state-of-the-art facilities of the packaging machinery, microbial invasion is inevitable in most of the packaged food, which not only degrades the food but also can elicit the pathogenic attack to the consumers. Therefore, various such markers/indicators have been targeted to check the spoilage in the QCs at the warehouses and the point of use for safer food consumption. These include pesticides, antibiotics, pathogenic microbial forms, allergens, heavy metal traces, etc. (Luong et al. 1997; Lozano et al. 2019). Pesticides like organophosphate, carbaryl, carbofuran, methomyl, Iprovalicarb, etc. are majorly used for agriculture-based food production, which eventually transfer to the crop due to their persistent accumulation in soil (Zamora-Sequeira et al. 2019). Although the antibiotics do not directly impact the health of the consumer in their prescribed dosage, their overdose can potentially harm the health by initiating side effects. Their uncontrolled use in the food industry to check microbial invasion in processed food to meet the prolonged shelf-life could trigger resistances against these, which eventually becomes a major threat to public health (Landers et al. 2012). The common antibiotics used in the food industries are penicillin, tetracyclines, and quinolones; however, these are not limited. (Landers et al. 2012). While various sterilization processes have been employed to get rid of microbial forms, their invasions are inevitable in the food, either fresh or processed ones. This is because of the available nutrients for their growth, which is an essential requirement. Mainly the bacterial, fungal, and viral-based pathogenic elements have been reported in food. Among all, a few have become threatful to individuals and public health concerns. Thus, for their determination in food, various microbe-specific determinants/markers are targeted to achieve food safety. Similarly in agriculture-based foods, various allergens and heavy metal traces are most commonly found in food items. Moreover, the adulteration in food also degrades its quality and sometimes becomes a health hazard.

11.3 Biosensors for Food Quality and Safety

The quality preservation of the food is important. For its assurance, several biosensor modules have been reported. This section discusses various biosensors developed that have the potential to be applied in the food industries.

11.3.1 Biosensing Prototypes for Supporting QC's of the Beverage Industry

The quality of beverages is governed by their constituents and ingredients. For example, in sweetened beverages, the sugar content is crucial for its taste and the health of the consumer. Excessive of it causes disorders such as obesity and diabetes, to the consumers. Thus, the detection of the glucose becomes crucial in the sweetened beverages (eg. juices, carbonated drinks, etc.) apart from its well established biomedical applications (Majer-Baranyi et al. 2008; Cinti et al. 2020; Kostejnova et al. 2021; Zhuang et al. 2021). The most common bioreceptors used for glucose sensors are glucose oxidase and glucose dehydrogenase. The diversity in the glucose biosensor development is due to its extremely stable enzyme bioreceptor "glucose oxidase," which offers a higher specificity, turnover rate, and greater stability. Another crucial parameter for the quality of the beverage is the fructose concentrations. The excessive consumption of it can cause serious health problems (i.e., fructosuria) in the individual's deficit of the fructokinase, which is the enzyme that breaks fructose. Therefore, fructose monitoring is required not only for beverage quality but also for consumer protection.

For developing the fructose biosensors, the d-Fructose-5- dehydrogenase and hexokinase coupled with fructose-6-phosphate kinase enzymes are commonly used.

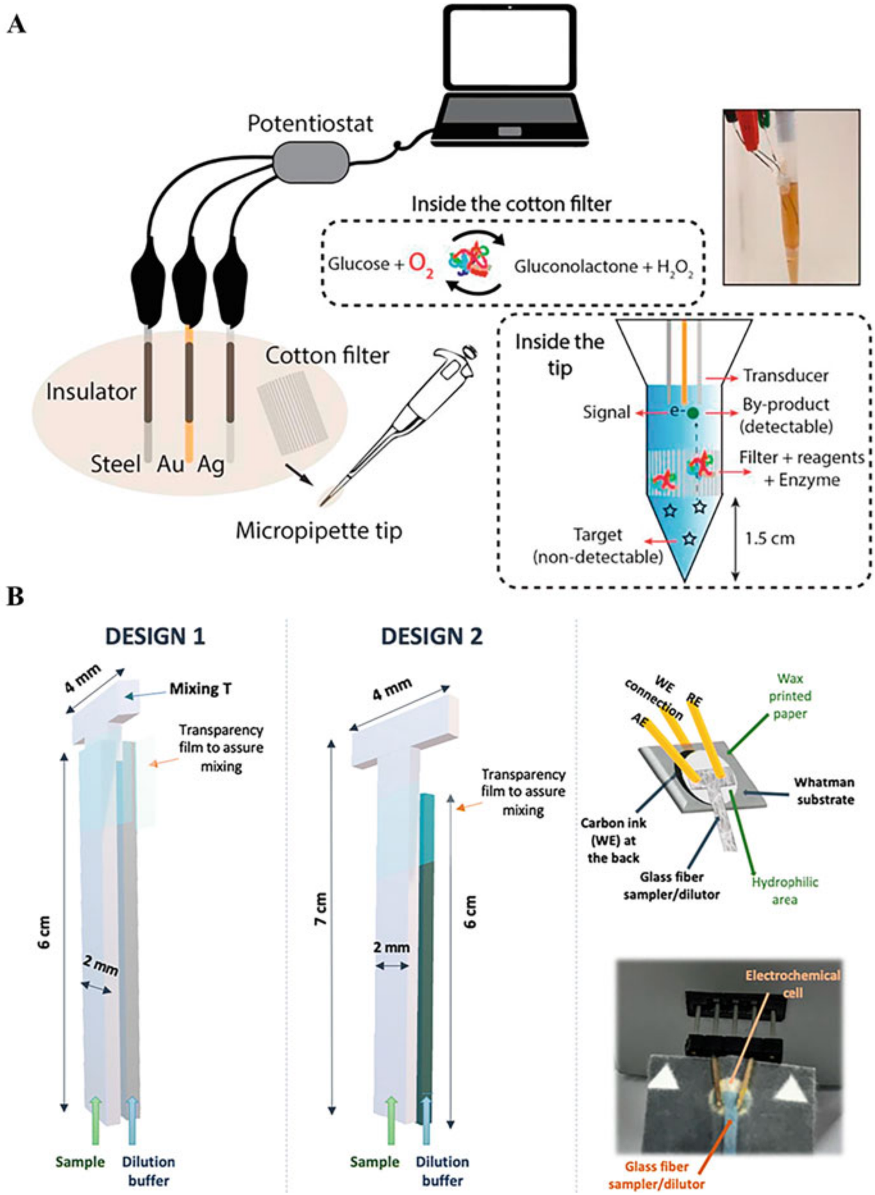

Fig. 11.2 Potential works that could serve the accurate detection of glucose contents in beverages: (**a**) working principle and the different parts of the lab-in-a-tip based approach by fusing wire electrodes into the pipette tip (Reprinted with the permission from Cinti et al. 2020; © Elsevier Inc.). (**b**) A paper assisted sampling of the beverage samples to the testing area for the detection of glucose using the wire-electrodes. (Reprinted with the permission from Amor-Gutiérrez et al. 2021; © Springer Nature)

In recent years, several biosensing prototypes for quality estimations have been developed for detecting the alcohol, sugar, vitamin, and other nutrients in alcoholic and non-alcoholic beverages. For instance, a pipette-based lab-on-a-tip electrochemical sensor (Fig. 11.2a) was reported for the rapid detection of glucose content in beverages with excellent analytical performance. The developed biosensor has shown a dynamic range of 0.5–10 mM with a detection limit of 170 μM (Cinti et al. 2020). Similarly, in another report, a glucose biosensor has been developed using a lab-on-paper approach for delivering a cheaper and more rapid detection of the sugar content in beverages (Fig. 11.2b) (Amor-Gutiérrez et al. 2021). This sensor is capable of detecting glucose in the concentration range between 0 mM and 15 mM in commercial orange juice and cola beverage samples. Using the same device, the authors have demonstrated the efficacy of the developed sensors for the detection of glucose from several samples simultaneously (Amor-Gutiérrez et al. 2019). Apart from the quality of beverages, safety and security are crucial for maintaining their quality for consumption, and thus the preservative, contaminants, and spoiling agents were also attempted to detect. Although, in alcoholic beverages, the microbe-borne spoilage is mostly unlikely, sugary non-carbonated beverages are more prone to such invasions. Thus, normal and pathogenic bacteria detection is routinely required. In addition, allergens and toxins are other more common threats found in beverages. Recent developments have targeted the detection of all such safety threats (Goud et al. 2016, 2017, 2019). For instance, Goud et al. have developed a miniaturized biosensor for the detection of aflatoxin B1 in alcoholic beverages. The sensor has shown remarkable analytical performance with the dynamic range and detection limits 0.05–6.0 ng mL^{-1} and 0.05 ng mL^{-1}, respectively (Goud et al. 2017).

11.3.2 Biosensing Prototypes for Supporting QC's of the Milk Industry

Another major segment of the food industry is based on milk and its products. The milk is enriched with various compounds including lactose, fat, citrate, nitrogen, casein, minerals, and non-proteinaceous nitrogen compounds that make milk a staple food. Lactose sugar in the milk is an indicator of its quality and the products. It also serves as an indicator of mastitis, where its level decreases upon the progression of the disease. Also, at the QCs, lactose estimation becomes more important due to its intolerance among the consumers. Several biosensing prototypes have been developed for the detection of lactose in milk. For instance, an enzymatic approach has been employed to develop the milk assessment, where β-galactosidase and glucose oxidase were co-immobilized on the sensor surface to develop the sensor probe. In the presence of lactose, the β-galactosidase enzyme converts it galactose and glucose eventually increasing the concentration of glucose. The generated glucose is subsequently cleaved by the co-immobilized glucose oxidase enzyme. This sensing prototype has shown a lower detection limit for lactose detection and was reported with 0.17 mg mL^{-1}(Jasti et al. 2014). Similarly, another device is developed by

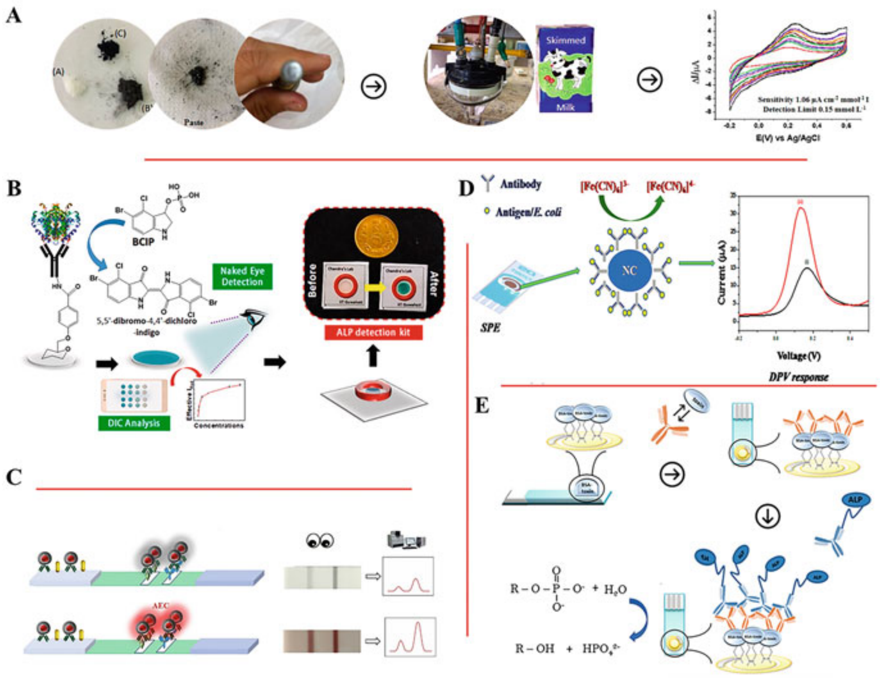

Fig. 11.3 Potential reported works that could lead to milk safety and quality improvement in dairy and dairy products: (**a**) Sensor prototypes for quality assessments based on lactose estimation in dairy products. (Reprinted with permission from de Brito et al. 2021, © Elsevier Inc.). (**b**) Portable colorimetric device for the qualitative and semi-quantitative estimation of milk pasteurization based on alkaline phosphatase content. (Reprinted with permission from Mahato et al. 2019, © Elsevier Inc.) (**c**) Lateral flow–based device for milk safety assessment based on the antibiotic (streptomycin) detection. (Reprinted with permission from Wei et al. 2020, © Elsevier Inc.) (**d**) Sensitive electrochemical device for the detection of bacterial forms *(Escherichia coli.)* in milk and milk products. (Reprinted with permission from Khan et al. 2021, © American Chemical Society). (**e**) Immunosensor-based chip for onsite detection of toxins (aflatoxin M1) in milk. (Reprinted with the permission from Karczmarczyk et al. 2017, © Elsevier Inc.)

de Brito et al. by employing electrochemical biosensors using lactase enzymes in nanomaterial-based transducer materials (Fig. 11.3a). This biosensor has shown excellent analytical performance with the detection limit of 0.15 mmol L^{-1}. The operational stability of the sensor was reported, which was found to be 12 h in consecutive usage of 10 days. Due to the nutrient richness of milk, microbial invasions are most common, which eventually lead to its spoilage, thereby, spoiling the milk by producing lactic acid. The invasions could also harbor the pathogenic bacteria, and consumption of which could cause more severe infectious pathogenic diseases. Therefore, to avoid such milk-borne contamination, a pasteurization process is employed, which kills all microbial forms. For confirming pasteurization, alkaline phosphatase is used as an indicator. Owing to the catalytic properties, alkaline phosphatase cleaves the chromogenic substrate and produces colorimetric detection. Exploiting this behavior, Mahato et al. have developed a miniaturized device for the detection of alkaline phosphatase, thereby estimating the pasteurization status (Fig. 11.3b) (Mahato and Chandra 2019). The lower concentrations (in milli-units per liter) confirm the higher degree of pasteurization (ideally the pasteurized milk contains <250 mU of the alkaline phosphatase enzyme) as most of the molecules of it get denatured at this temperature. This device can detect alkaline phosphatase in the range of 10–1000 U/mL, which covers the entire concentration ranges of raw milk from a healthy cow and mastitis cow. This device has shown an excellent capability of sensing the target qualitatively and quantitatively when coupled to the smartphone. For achieving better productivity, antibiotics are commonly being employed for livestocks growth and commercial rearing. A study reports, approximately 63.151 (± 1.560) tons of antibiotics are being employed in livestocks yearly. The milk produced from such dosed livestock has found a significant amount of antibiotics due to their injudicious usage. Although antibiotics do not directly harm the consumers, the microbiota inside the gut may develop resistance on their constant exposure, which can be lethal if any virulency occurs inside the gut. To check the antibiotic contaminant in milk, several biosensors have been developed, using colorimetric and electrochemical formats. For instance, Wei et al., have developed lateral flow assay-based colorimetric detection of streptomycin using the gold–platinum bimetallic enzyme incorporated with a tetramethylbenzidine/hydrogen peroxide-based colorimetric system (Fig. 11.3c). This has shown a reasonably great analytic performance, where the detection limit of 1 ng mL^{-1} in the lateral flow module. Such efficient detection methodologies can be adopted for the onsite detection of various other antibiotics. The microbial invasions can also be tested by checking their populations in milk, however, the direct counting is hectic and laborious. Thus, biosensing strategies have been employed to detect the bacterial concentrations in milk. In this context, several works have been reported, which directly (label-free) or indirectly (with label) assess the presence of bacteria in milk. In this context, Khan et al. has developed a biosensing module for the detection of *Escherichia coli* from milk samples. The sensor was developed using the nano empowered miniaturized electrochemical transducing system, which has offered better sensitivity (Fig. 11.3d). The developed sensors show the detection limit of 2 CFU mL^{-1} using a redox couple. Toxins are the other contaminating agents, produced by the microbial forms. Consumption of toxins can cause severe complications in an individual's health. In contaminated/spoiled milk, several such toxins have been reported including, 5-vinyl-oxazolidine-2-thione, pyrrolizidine, swainsonine, trematode, aflatoxin M1, etc. (Liener and Liener Bsc 2002). Biosensors have been developed for the detection of such toxins to check the suitability of the milk for consumption. For instance, an immunosensor has been developed for the detection of Aflatoxin M1 (Fig. 11.3e). The sensor probe has been designed using the capture antibody and the secondary antibody conjugated alkaline phosphatase on the modified gold screen printed electrodes.

11.3.3 Biosensing Prototypes for Supporting QC's of the Meat Industry

Another major food sector is the meat industry, where various kinds of meat produced from livestock are processed, packaged, and commercialized. For producing good quality meat, the rearing of the livestock and storage conditions play an important role. Good and hygienically reared livestock produce healthy consumable meat that is devoid of pathogens, pesticides, drugs, heavy metals, and toxins, which are regulated by the regulatory bodies for commercialization to ensure good-quality meat for safer consumption. Although strict laws and consumer awareness have helped to maintain the quality of commercial meat to a greater extent, the inevitable malpractices even on small scale can spoil the product of the entire batch. In addition, the integrity of meat is also a big concern in the global society. To maintain integrity, various regulatory bodies are functioning across the world, which ensure and certify for purity by identifying the mixtures of meat. The conventional instruments for their detections are highly sensitive and are capable of detecting such contaminants in meat. However, employing these for the detection of every retailer/consumer is not a practical solution. This limits the usage of instruments to save the consumers from consuming spoiled meat even after having a great analytical performance. Thus to ensure safe consumption, onsite detection is essential.

So far, several miniaturized modules based on biosensors have been reported for the determination of meat quality. In a report by Labrador et al., a biopsy needle-based sensor has been developed, which can sense nitrate, nitrite, and NaCl contents in the meat samples, which are used as preservatives and carry carcinogenic effects. This biosensor has been tested in a real meat sample, and showed excellent profiling of nitrate and nitrite detection (Fig. 11.4a). Similarly, nitrates present in the meat have been tested using an electrochemical method. The sensor probe was developed using mesoporous carbon composite materials to obtain sensitive detection. The detection showed excellent analytical performance with the detection limit of 2.1 nM. Another major challenge is to keep the integrity of the meat quality in terms of the source of meat. In this context, Flauzino et al. have developed an electrochemical module for the detection of meat adulteration (Fig. 11.4b). The sensor detects the specific DNA sequence of porcine mitochondrial origin to find meat adulteration. The developed sensor shows 45 days of stability with the detection limit of 9% by the weight corresponding to beef and pork weight. Similarly, targeting porcine mitochondrial DNA, Ali et al. have developed a sensitive biosensor based on carbon-reduced graphene oxide electrodes using the screen-printed electrode module (Hartati et al. 2020). The excellent analytical performance dynamic range of 0–10 μg/mL and the detection limit of the 1.76 μg/mL of target DNA are capable of detecting the meat integrity in many commercial contexts.

In addition, antibiotics such as chloramphenicols, tetracyclines are being extensively used in the rearing farms from the birth of the livestock, which has become a food security hazard. Other chemicals such as ractopamine and clenbuterol are commonly used with feed to obtain increased growth and lean meat of the animals, which is a threat to the human cardiovascular and nervous systems if consumed for a

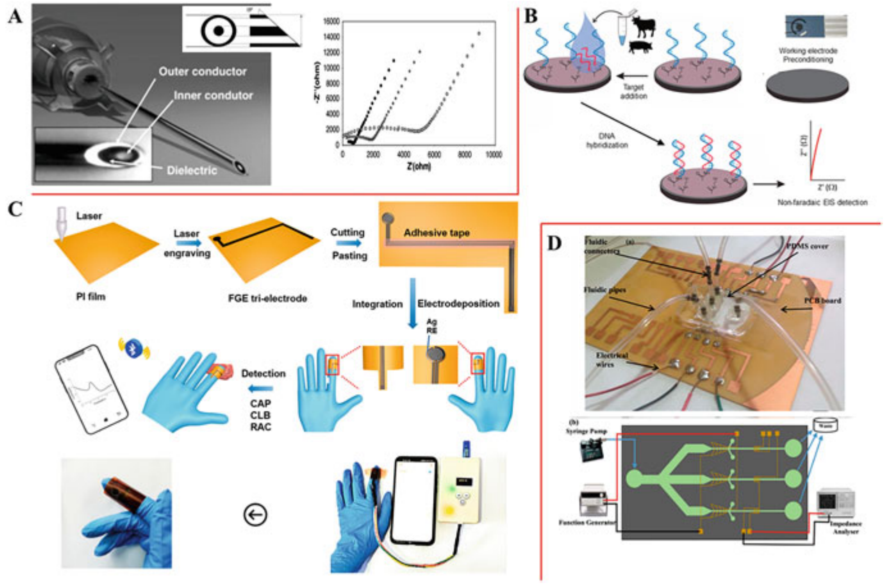

Fig. 11.4 Potential works that could lead to meat safety and quality improvement in the meat industry and products: (**a**) Portable needle-based device for nitrate, nitrite, and NaCl detection in meat samples for estimating the meat quality. (Reprinted with the permission from Labrador et al. 2010, © Elsevier Inc.) (**b**) Device for meat adulteration safety and integrity assessment based on the gene identification. (Reprinted with permission from Flauzino et al. 2022, © Elsevier Inc.) (**c**) Flexible electrochemical device for meat-based fast food security based on antibiotic profiles (chloramphenicol, clenbuterol, and ractopamine) detection. (Reprinted with permission from Li et al. 2022, © Elsevier Inc.) (**d**) Immunosensor based device for the onsite detection of *Salmonella serogroups* in meat samples. (Reprinted with the permission from Jasim et al. 2019, © Elsevier Inc.)

prolonged duration. Considering the potential health hazards, these drugs are banned in many countries across the globe. To detect such drugs in meat, Li et al. have developed a flexible globe-based electrochemical biosensor for the simultaneous detection of chloramphenicol, clenbuterol, and ractopamine in the meat sample (Fig. 11.4c). The developed sensor shows excellent analytical performance where the detection limits of 2.70, 1.29, and 7.81 μM and linear ranges of 10–200, 5–80, and 25–250 μM were obtained for chloramphenicol, clenbuterol, and ractopamine, respectively. Microbial invasions are another factor for meat spoilage and thus the detection of such contaminants and spoiling agents are of utmost need not only for the prolonged shelf-life of the meat but also to save the consumers from pathogens. In this context, an attempt has been made by Jasim et al. where they have developed microfluidic-based immunosensors for the detection of *Salmonella* serogroups of bacteria (Fig. 11.4d) using the electrochemical impedance spectroscopy. This biosensor is capable of delivering an excellent limit of detection of 7 cells/mL. The entire detection time for this sensor is approximately 40 minutes, which is a little longer for onsite detections. The developed biosensor could serve as the potential module for bacterial detection in meat samples in onsite settings if the detection time is improved.

11.4 Commercial Biosensors for Food Quality Assessment

The important requirement of onsite QCs is the complete profiling of the food items, which has attracted simultaneous assessment of the given sample for delivering efficient controlling of the quality. These include biosensor contaminants, sugar contents, alcohols, amino acids, flavors, sweeteners, etc. Also, food allergens, toxins, pathogens, and additives are the major targets for these commercial devices. In the demand for these, the commercial instruments incorporated with multiple biosensors have become more promising to the food industries. The YSI 2700 Select food analyzer is the most prominent biosensors-based commercial instrument, which can detect essential amino acids, lactose, glucose, ethanol, and starch simultaneously. Thus, this instrument has been in demand at the QCs of several food processing industries (viz. beverages, meat, dairy, etc.). Similarly, another instrument, ABD 3000 biosensor assay system, is a multiplexed biosensor array that can detect and quantify L-lysine alcohol, L-amino acids, ascorbate, glucose, lactate, lactose/galactose, oxalate, and sucrose in the sample. These modules are employed for food safety, however, their cost-consuming nature practically limits to be used for every sample. Thus, to realize the onsite QCs for food products, a cheaper alternative is required to minimize the burden on the QC-centers for quality assessment as well as in the points of retail. The common formats of commercial devices are integrated autoanalyzer, manual benchtop, and most advanced portable biosensors. Few of them are reported in the table below (Table 11.1) (Bahadir and Sezgintürk 2015). The commercial success of these spin-offs from the industry standards' customized products indicates the competency for safeguarding the food products for consumption.

11.5 (Bio)/Sensors for Food Packaging

Several smart strategies have been adopted to protect the quality and prevent food spoilage by using biosensors. Among all, smart packaging strategies with biosensing modules have found great attention in recent times (Ghaani et al. 2016). These active packages when exposed to the environment of potential indicators, issue an alert. Although such biosensor-based active food packaging cannot detect the quality or the spoilage of the packaged food item, it can certainly be able to detect the possible exposure of contaminating and spoiling agents from the environment, which would prevent mass spoilage (Sobhan et al. 2021). In recent advancements, these are being coupled with artificially intelligent machinery and spatiotemporal geotagging facilities, which makes them commercially viable. These strategies are not only capable of avoiding mass spoilage but also offer an increased shelf-life. Mainly, these smart packaging systems follow the purpose of "something extra," which is a

Table 11.1 Various commercial food sensors have been summarized (Copyright 2015; reused with the permission of Bahadir and Sezgintürk (2015))

Company	Biosensor	Country
Oriental Electric	Fish deterioration tracking	China
Massachusetts Institute of Technology	Detection of E. coli O157:H7 in lettuce (canary)	USA
Michigan State University's electrochemical biosensor	Detection of E. coli O157:H7 and salmonella in meat products in the USA	USA
Georgia Research Tech Institute	Detection of salmonella and campylobacter in the pork industry	USA
Naval Research Laboratory	Detection of staphylococcal enterotoxin B and botulinum toxin A in tomatoes, sweet corn, beans, and mushrooms	USA
Universitat Autonoma de Barcelona in collaboration with CSIC	Detection of atrazine traces	Spain
Molecular Circuitry, Inc.	E. coli O157, salmonella, listeria, and campylobacter	USA
Research International	Proteins, toxins, virus, bacteria, spores, and fungi (simultaneous analysis)	USA
Universal Sensors	Ethanol, methanol, glucose, sucrose, lactose, l-AAs, glutamine, ascorbic acid, and oxalate	USA
Texas Instruments, Inc.	Peanut allergens, antibiotics	USA
Yellow SPRINGS Instruments	Glucose, sucrose, lactose, l-lactate, galactose, l-glutamate, ethanol, H2O2, starch, glutamine, choline	USA
Affinity sensors	Staphylococcus aureus and cholera toxin	UK
Ambri Ltd	Pathogens such as salmonella and enterococcus	USA
Biacore AB	Water-soluble vitamins, chemical veterinary residues, and mycotoxins	Sweden
BioFuture Srl	Glucose, fructose, malic acid, and lactic acid (fermentation)	Italy
Biomerieux	Microorganisms	France
Biosensor systems design	Microorganisms and toxic substances	USA
Biosensores S.L.	Toxic substances	Spain
Chemel AB	Glucose, saccharose, ethanol, methanol, and lactose	Sweden
IVA co. Ltd	Heavy metals	Russia
Motorola	Microorganisms and genetically modified organisms	Japan
Iventus Bio Tec	Ascorbic acid	Germany
Analox Instruments	Ethanol, methanol, glucose, lactate, glycerol	UK, USA
Gwent Sensors	Glucose	UK

fundamentally valued addition to the batch of products. The common parameters that are crucial for any food storage and its prolonged shelf life are the pH, temperature, and moisture contents, which are directly related to its freshness. Thus, the smart sensor–based packaging mainly contains the modules that primarily detected the above parameters. The commercially available smart packaging modules have mainly included gas indicators (Soon and Manning 2019), time–temperature indicators (Mohebi and Marquez 2014), freshness indicators (pH sensor, metabolite sensors) (Fang et al. 2017), pathogens (toxins, bacterial, and fungal indicators) biosensors (Fang et al. 2017), etc. The technological advancement in communication has supplemented the tracking features, that have been exploited in building these strategies, where the indicator mechanism has been coupled with the communication module with barcode-enabled RFID patches for easy screening.

These are various opportunities for biosensors in the packaging and handling of food. Mainly, these include food freshness, food integrity, fruit ripening, contamination, spoilage detection of food, and commercialized food items, especially where the food is preserved for a long time. The freshness of protein-based food is a common concern and most of them are prone to bacterial or fungal invasions. The spoilage of them produces nitrogen-based compounds (viz. ammonium gas, nitrogen gas, aldehydes, ketones, etc.) that can be detected by the colorimetric biosensors. For instance, an indirect biosensor (using the glucose-sensing strategy), proposed by smiddy et al. detects the L-cysteine which is a biomarker of meat freshness and its spoilage (Smiddy et al. 2002). The phenolic compounds are majorly released in ripening fruits, and thus sensing these indicators can tell the ripening stages and the spoilage by overripening. For example, malic acid has been developed for the detection of fruit ripeness (Vargas et al. 2016). Additionally, in the most ripened fruits, the sugar content increases over-ripening, which can also be exploited in the onsite QCs while packaging the batch of fruits. The growing needs for the food items have attracted the overuse of pesticides and antibiotics in crop production, which eventually get stored in the food items, and may be harmful if consumed above the permissible limit. Thus, biosensors detecting such analytes can be incorporated in smart packaging in a comprehensive examination of the packaged food at the QCs and packaging centers. The conceptualization of smart and active packaging has been since the early 2010s; however, these have been limited to get commercial attention due to their technical challenges, which include integration difficulty, miniaturization capabilities, accuracy, and the cost of fabrication. However, in recent decades, the advancement of microfabrication process and the nano empowered techniques have significantly increased the accuracy and lowered the cost of the biosensing modules, which may play a key role for realizing smart packaging for saving the food.

11.6 Conclusions and Future Directions

Healthy food is an essential need for good health. However, the malpractices have severely compromised the quality of food to a greater extent. At the commercial level, various technologies including intelligent packaging, safety, and cold storage transportation have been supported to retain the food qualities; however, microbial invasion and deliberate adulteration have attracted serious concerns. The biggest challenge in food industries is the preservatives, which provide a longer shelf-life; however, a few of them prove severely detrimental if consumed above the limit. The onsite detection of those is the best way to prevent toxic-level consumption. This chapter summarized the various attempts taken for saving food by retaining its qualities using the biosensing modules. The food biomarkers commonly targeted for ensuring the quality and detection of spoilage have also been discussed. Here, the focus was to collate the available prototypes that could be employed at existing QCs or at the point of need for facilitating the efficient screening of food for safer consumption. The future direction would be to discover customized exclusive biomarkers for a particular food and the development of low-cost biosensing modules for it to save the consumers from health deterioration and mass spoilage of food during its commercialization and storage.

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