

1. Overview

The dimension of food integrity widened after many food scandals emerged, not only focusing on the scope of nature, substance, and quality and safety of food but also concerning the food production, including “the way it has been sourced and distributed and being honest about those areas to consumers” (Elliot, 2014). Among the incidents, the horsemeat scandal in Europe, the contamination of melamine in infant formula milk in China, and the mislabeling of fish species in the United States hit the sensation of food integrity. The main interest of food integrity is in food supply chain management. However, it can be categorized into developing quality management, food safety management systems, and food integrity management systems (Manning, 2017). For instance, developing food integrity management systems into the broader food supply chain aimed firstly to guarantee safety, quality, and authenticity; secondly to ensure reliable labelling, and thirdly to ensure effective management of provenance such as halal status (Manning, 2017). After all, the main elements of food integrity should be concerned with product, process, people, and data integrity. The example of each component in food integrity has been tabulated in Figure 1.

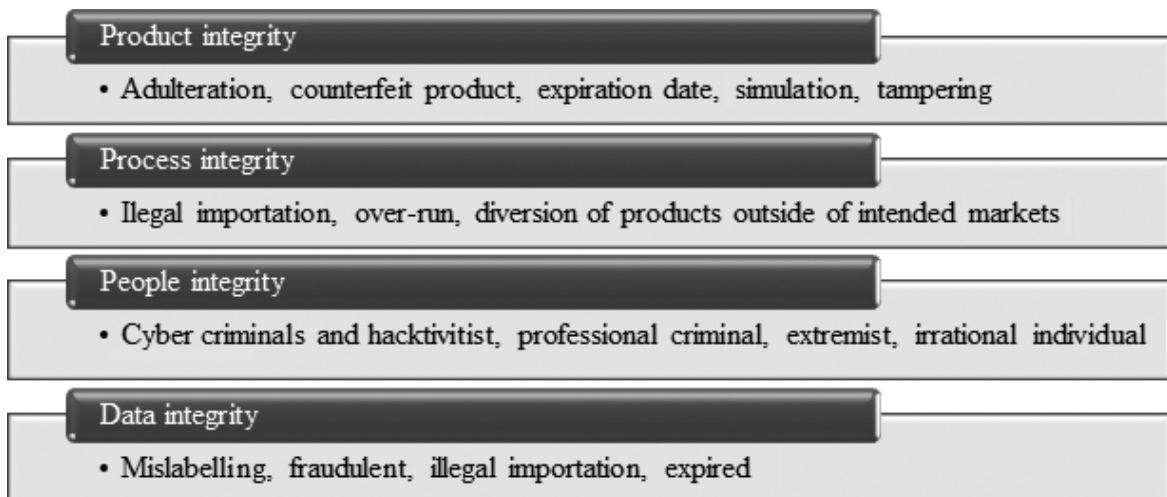


Fig. 1 Examples of each element of food integrity (Manning, 2017)

2. Food product integrity

Food integrity denotes that all people can have access to safe, authentic, and nutritious food at all times. In this chapter, the focus is given on the role of food science and technology in verifying food product integrity. Food integrity consists of several elements: quality, safety, authenticity, traceability, and adulteration.

Food quality can be measured based on the food characteristics acceptable to the consumer. The physical characteristics of food include texture, flavor, and appearance of food products such as size, shape, color, and grade. Chemical characteristics are related to the nutritional content of

the food product. In addition, the high quality of food that meets the standards set up in legislation and product specifications is also related to ethical and sustainable production concerning the consumer's acceptability.

Food safety can be defined as the manufacturing practice of preparing, handling, producing, and storing food to avoid contamination and maintain the nutritional value of food in supporting a healthy diet (FAO, 2004; Sheikha, 2015). Any biological, chemical or physical agent that can cause any illness or injury without control is considered a food hazard. Biological hazards that can result on the foodborne illness outbreaks are the major concern in the food industry. The common organisms that can cause infection, intoxication, and even death include *Salmonella*, *E. coli*, and *Clostridium botulinum*. Chemical agents such as pesticides and heavy metals that can cause hazards in food can be categorized into three groups: (i) natural chemicals; (ii) chemicals used in formulation for food products, or (iii) chemicals that are unintentionally or incidentally present in the finished product. Physical agents that can cause physical hazards are not limited to bone, glass, wood, faces, etc. Therefore, risk assessments and suitable mitigation measurements can be carried out based on the research and previous data on food safety hazards.

Generally, the purpose of food fraud is to gain financially by the enhancement of the apparent value of the product or reducing the production cost. It can be classified as adulteration, tampering, product overrun, diversion, theft, and counterfeiting. The adulteration or contamination of foods is considered if the valuable component in the food is removed or replaced with a cheaper or poor quality to concealed with an actual food product or substitutes instead of expensive ingredients (Goyal et al., 2022; Roy & Yadav, 2022). Food products can be easily adulterated since it has a lengthy, complicated processing and supply chain. Therefore, food authentication is important to ensure the safety and quality of food products and consumer protection. It is also important for both official bodies and industries in charge of labelling and raw materials and finished products to be tested for compliance with the national legislation, international standards, and other guidelines and specifications (Baeten & Dardenne, 2002).

The high food adulteration incidence was reported in the literature and can be categorized into six groups which were '*animal origin and seafood*' for milk and dairy products, meat and meat products, fish and seafood; '*oils and fats*'; '*beverages*' for fruit juices, coffee and tea, alcoholic beverages; '*spices and sweeteners*' for spices and extracts, sweeteners including honey; '*grain-based food*' for cereals and pulse; and '*others*' for organic foods and dietary supplements (Hong et al., 2017). The key players, including the food industry, academia, and government, are aware of this serious issue and have implemented efforts to ensure food integrity, including methods and procedures. A competent, fast, and reliable analysis is needed to overcome the challenges and ensure product quality. The authentication and adulteration of food can be validated by various technique including physical, chemical/ biochemical, or DNA molecular depending on the type of adulterant to be detected (Bansal et al., 2017). This use of analytical methods such as non-destructive radiative evaluation, fingerprinting characterization, novel technology for rapid detection, and authentication were reviewed.

3. Physical technique

The authenticity of food products can be examined by analyzing the physical characteristics of food such as color, texture, solubility, bulk density, morphological traits, etc (Bansal et al., 2017). Physical techniques such as microscopic and macroscopic can be used for the physical characterization.

In the early years, Louveaux et al., (1978) detected honey adulteration with cane sugar and cane sugar products through optical microscopy. Combining microscopy and macroscopy with chemical profiling also identifies and authenticates herbs and medicinal plants (Sheorey & Tiwari, 2011). The utilization of microscopy and macroscopy helps detect microbial contamination in food such as fungi. Furthermore, the botanical origin can be detected with the advent of electron microscopy. For instance, the origin of honey can be determined by analyzing the surface pattern of pollen from honey (Jones & Bryant, 2014).

4. Chemical/biochemical technique

In general, the chemical/biochemical techniques offer more accurate results, and the food's contaminants can be detected even at lower concentrations. However, these techniques are costly, time-consuming and require well-trained operators to conduct the experimental works. The chemical/biochemical techniques can be categorized into four main categories: (1) spectroscopic-based, (2) chromatographic-based, (3) immunologic-based, and (4) electrophoretic-based techniques. The focus of this section is given to the spectroscopic-based and chromatographic-based technique since these two techniques are the most used especially in food adulteration and authenticity.

4.1 Vibrational spectroscopic techniques

A range of vibrational spectroscopic techniques such as Fourier transform near-infrared spectroscopy (FT-NIR), mid-infrared (MIR), Raman spectroscopy, and hyperspectral imaging (HIS) has been successfully used as sensitive and fast analytical techniques for the determination of food contamination. Figure 2 shows the typical vibrational spectroscopic techniques used for food authentication. The main advantages of these techniques are being non-destructive, relatively low analysis cost, being adopted for both qualitative and quantitative analysis, and providing an alternative to wet-chemical and time-consuming techniques (Lohumi et al., 2013).

NIR region is between 780 to 2500 nm and can detect overtone and combination bands for the chemical bonds between light atoms, such as C-H, O-H, and N-H that have high vibrational frequencies (Osborne et al., 1993). FT-NIR can be used for qualitative and quantitative analysis with little or no sample preparation (Lohumi et al., 2015). Continuous development makes this technique reliable and efficient, and it has become a standard analytical method for quality control measurements. FT-NIR can be categorized into three groups based on instrumental devices: (i) sequential instruments - the absorbance is sequentially collected in time, and the instrument

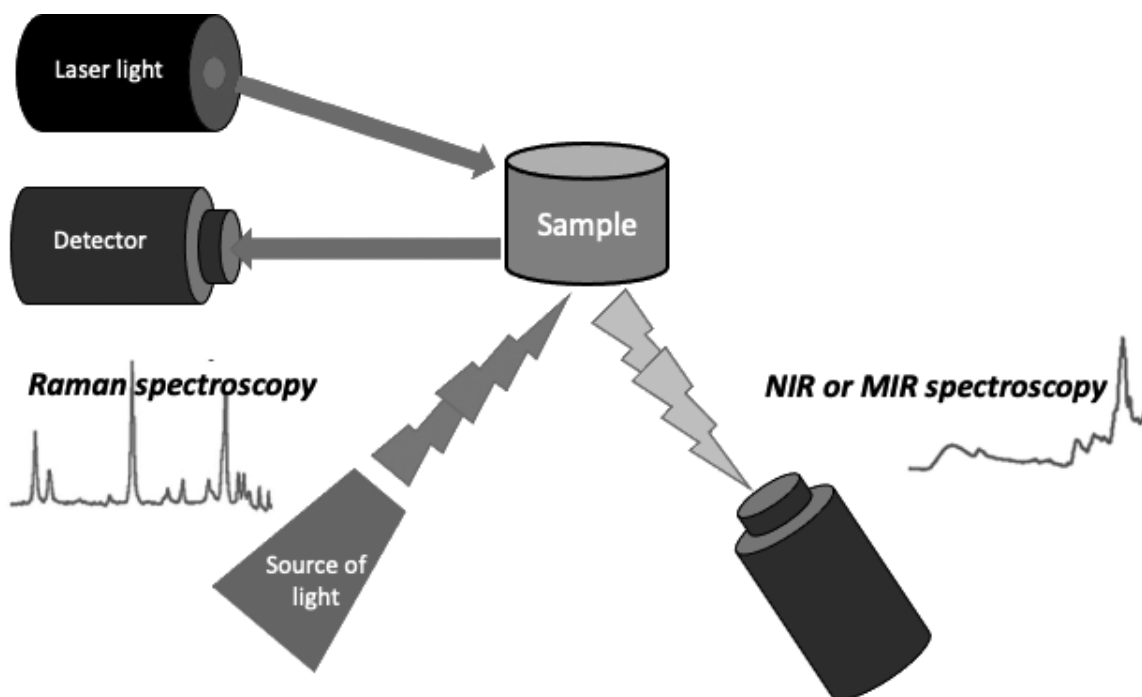


Fig. 2 Schematic diagram of typical vibrational spectroscopic techniques.

is equipped with a monochromator or filters; (ii) Fourier transform or multiplex instruments - several frequencies are detected simultaneously in the form of an interferogram; and (iii) multi-channel instruments - several detectors separately detect the absorbance at several wavelengths (Lohumi et al., 2015).

Numerous studies on detecting food adulterations have been conducted using this technique. For instance, identification of animal meat muscles (Cozzolino & Murray, 2004), detection of beef hamburger adulteration (Ding & Xu, 2000), adulteration in crabmeat and species authenticity (Gayo et al., 2006). Besides that, this technique also successfully detected protein adulteration in yogurt and melamine contamination in milk powder (Haughey et al., 2013) and soya bean meal (Xu et al., 2013). NIR spectroscopy can be used in different spectral modes, including reflectance, transmission, interactance, and transflectance, to estimate the sample's external and internal properties. NIR reflectance spectra (1100–2498 nm) managed to detect 10% pulp wash in orange juice and sugar-acid mixtures with 90% accuracy (Downey et al., 2016). The transmission mode offers the most straightforward sampling technique for analyzing solid, liquid, and gas samples. It can be used to detect a sample's external and internal qualities. For example, this mode has been used to differentiate white wine based on the origin (Daniel Cozzolino et al., 2003) and detection of fatty acid composition in beef and chicken (Riovanto et al., 2012). The interactance mode is used for the internal information of a sample when the transmission measurements are challenging to obtain. The transflectance mode is the combination of transmission and reflectance modes. It is specifically designed to measure the spectra of thin or clear sample.

FT-IR focuses on the MIR region between 4000 to 400 cm^{-1} of the electromagnetic spectrum. It can produce a chemical profile of the sample through the fundamental vibrational and rotational stretching modes of molecules. FT-IR provides more excellent chemical information of the

sample compared to NIR spectroscopy because it measures fundamental vibrations instead of the overtones and combinations band measured in the NIR region (Lohumi et al., 2015). The MIR region can be divided into two, which known as the functional group region (4000 to 1500 cm^{-1}) and the fingerprint region (1500 to 500 cm^{-1}).

FT-IR also enables the measurement of all types of samples by using different measurement modes such as attenuated total reflectance (ATR), diffuse reflectance, high-throughput transmission (HTT), and transmission cell. ATR-FT-IR has been widely used in the analysis of food authentication because it can analyze both qualitative and quantitative analysis. ATR demonstrated a better sampling method with higher accuracy than HTT for a sampling technique such as microbial spoilage in milk (Nicolaou & Goodacre, 2008). Various combinations of FT-IR with sampling techniques have been successfully applied for adulteration detection in honey. FT-IR spectroscopy has also been combined with chemometrics to analyze high-value edible oils such as olive oil, avocado oil, etc. The adulteration in edible oils can be detected through a distinct classification due to the difference in fatty acid and triglyceride content (Christopoulou et al., 2004). The determination of sugar concentration in fruit juice also can be analyzed using FT-IR spectroscopy at the fingerprint region of MIR because this region has high sensitivity spectral region as compared to NIR. This suggested FT-IR is a versatile and powerful analytical method for food applications. FT-IR and NIR are compliments to each other with advantages and disadvantages for each technique.

4.2 Chromatographic techniques

Among fingerprint techniques, chromatography is found to be more informative in fingerprint analysis. The chromatographic method will provide information by translating the peak intensity presenting the area or height and their position (retention time). This method is convenient and reliable to characterize the food sample particularly for the used in identifying food authenticity. This is because, this chromatographic technique promotes vast and remarkable capabilities with high sensitivity, reproducibility, and robustness that enable them to provide the highest information content of the food products (Esteki et al., 2018).

Previously, using various types of detectors, the chromatographic technique has been used to identify chemical adulteration markers such as fatty acids, oligosaccharides, and tocopherols (Fanali et al., 2016). Most HPLC or GC methods were developed to detect carbohydrates, carotenoids, amino acids, phenolic or other organic compounds (Herrero et al., 2017). For instance, the chromatographic technique could detect the adulteration in oil and characterize the blend's composition of oils by determining triacylglycerols (TAGs), sterols, and fatty acids compounds present in the oil (Wernig et al., 2018).

The most widely used in detecting the adulteration or identification of the authenticity of food products are gas chromatography (GC) and high-performance liquid chromatography (HPLC). GC-FID is a popular chromatography mode due to being cheap, well-established, robust, and rapid. GC method was developed to detect adulteration in olive oil by comparing the fatty acid composition and chemometric analysis. GC method can be used to characterize and differentiate different vegetable oils by focusing on the fatty acid methyl esters (FAME), obtained

through trans-esterification of vegetable oils (Brodnjak-Vončina et al., 2005). For instance, the identification of extra virgin olive oil, which is an adulterer with corn, peanut, rapeseed, and sunflower oils, was successfully demonstrated by Yang et al. (2013) using the GC-MS method. GC with surface acoustic wave detector (GC-SAW system) combined with chemometrics was also used to identify the presence of lard in virgin coconut oil (Mansor et al., 2011). Besides that, the adulteration of extra virgin olive oil with 5% soybean oil has also been identified using the GC method by analyzing the fatty acids composition as an indicator of purity and the linoleic acid content as a parameter for the detection (Jabeur et al., 2014). The adulteration of almond powder with the apricot kernel was also detected using GC fatty acid fingerprinting combined with chemometric methods (Esteki et al., 2017). Nevertheless, frequently, GC needed derivatization to become the main limitation of GC.

This limitation can be overcome by using HPLC, which does not need any derivatization and can analyze both polar and non-polar compounds. HPLC has successfully demonstrated the adulteration of ovine, caprine cheese, and milk with bovine milk at lower levels (2 % v/v) by analyzing the whey protein β -lactoglobulin (Chen et al., 2004). Veloso et al. (2002) also detected bovine, ovine, and caprine milk using reversed-phased HPLC. HPLC method managed to classify wines from different denominations of origin in the Canary Islands, Spain, by analyzing 15 polyphenol concentrations combined with principal component analysis (PCA) and linear discriminant analysis (LDA) (Rodríguez-Delgado et al., 2002). The combination of PCA and LDA, as well as HPLC equipped with a UV detector, was also used to classify some Australian wines based on geographical groups with accuracy in the 75 to 100 % range (Bellomarino et al., 2009). In addition, reversed-phased HPLC combined with PCA and LDA was used to discriminate different varieties of green and roasted coffee beans by analyzing the triglyceride and tocopherol composition (González et al., 2001). The current trend shifting from HPLC to UHPLC due to the particle size of the stationary phase, sample size, and mobile phase have been reduced. Hence, it becomes more efficient, with high selectivity, throughput, speed, and resolution.

4.3 Electric nose

Most detection methods for food authentication are time-consuming and costly and require professional operators to conduct the analysis. As the adulteration scandals become intense recently, a real-time rapid and reliable detection method is needed to tackle this issue. This rapid detection technique can be used for commercialized processed foods in the market. One of the technologies invented is the electric nose (e-nose), which mimics the human nose as an artificial olfactory system or machine olfaction for automated simulation of the sense of smell (Roy & Yadav, 2022).

E-nose is classified as a non-destructive method widely used in many applications, including environment monitoring, quality control assessment in agricultural goods, fraudulent adulteration, and spoilages. This method can differentiate the quality of food products by detecting the specific components of an odor and analyzing its chemical makeup to identify it. This detection is preferred to routine laboratory analysis because of easy handling of the system, cost effective, and short time analysis (Roy & Yadav, 2022).

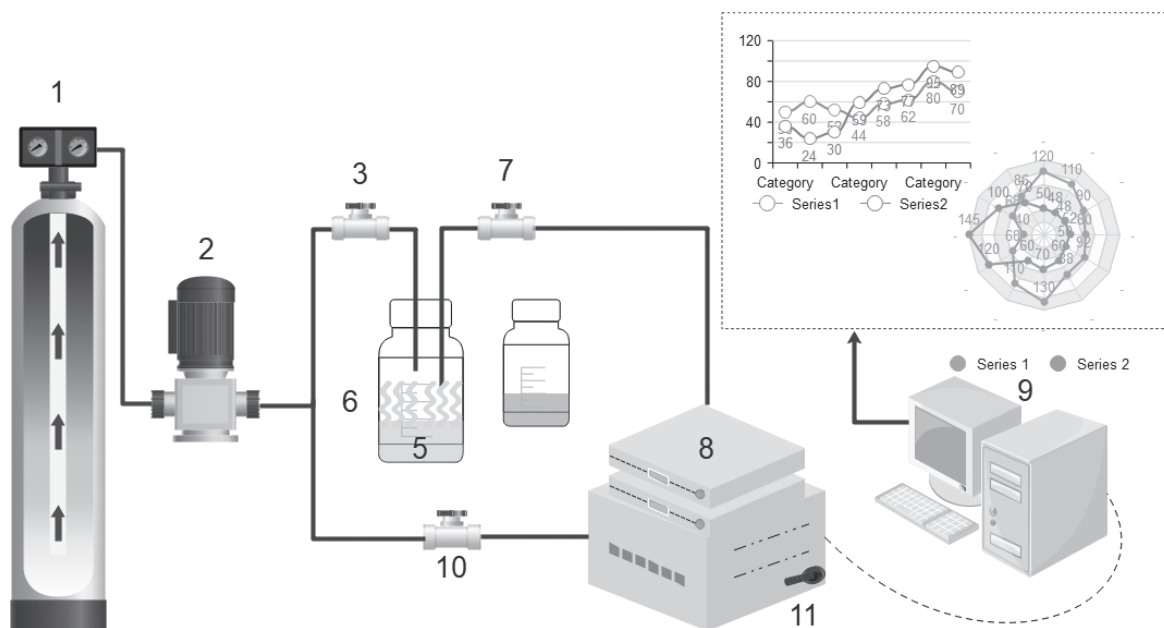


Fig. 3 The typical block diagram of an e-nose consists of: 1 synthetic air cylinder, 2 blower, 3 air inlet valve, 4 closed sample container, 5 sample, 6 volatiles, 7 sampling valve, 8 sensory array chamber, 9 data processing and pattern recognition, 10 purge valve, 11 purge outlet. (Source: Roy & Yadav, 2022).

Many research related to food quality using this e-nose has been reported previously. The idea of an e-nose system for aroma discrimination based on chemical sensors has been demonstrated by Persaud & Dodd (1982). In general, e-nose consists of four main units: (i) an electronic multi-sensor array system for chemical detection; (ii) an information processing unit; (iii) a digital pattern recognition algorithm system that is capable of recognizing odors; and (iv) a reference library database. Figure 3 illustrates the typical block diagram of an e-nose.

The mechanism of e-nose is based on the identification of volatile organic compounds generated from the food samples in the translation to voltage differences which are detected by limited selective sensors. Commonly used sensors are surface acoustic waves (SAW), and conducting polymer and metal oxide semiconductors (MOS). MOS sensor has been widely used because of their high chemical stability, low response to moisture, long life, and reasonable price. The synthetic air (99.9 % pure) blows the generated volatile organic compounds from the sample to the sensor array chamber. The air inlet and sampling valves remain open during the measurement, whilst the purging valve will be closed to introduce the generated volatile organic compounds to the sensor array chamber. The received analogue signal by the sensor array which consists of selective and sensitive gas sensors coupled with a data acquisition card will then convert to digital signals which can be read on the monitor (Roy & Yadav, 2022). In principle, food quality can be classified by differentiating the complex odors from food samples generated as voltage response value (output).

An e-nose has been widely used in detecting various food adulteration including honey, milk and dairy products, fruits, meat, fish, spice, edible oils, alcoholic and non-alcoholic drinks, tea and coffee, etc. The examples include the adulteration of edible plant oils with cheaper sources of oils, fats, and sterols, which have nearly similar fatty acid profiles. Sometimes, adulteration of edible

oils might cause serious health problems such as Spanish olive oil syndrome (Clemente & Cahoon, 2009). However, this e-nose technology is trustworthy and highly sensitive to ascertain the purity and authenticity of the edible oil. The authenticity of edible oils can be determined through various e-nose systems that have been developed. Hai & Wang (2006) detected the adulteration of Camellia seed oil with maize oil using an e-nose device with 10 MOS sensors. The adulteration of argan oil with sunflower oil using 5 MOS sensor-based e-nose (Bougrini et al., 2014). Besides that, Yu et al. (2007) also demonstrated the adulteration of skim milk with water and reconstituted milk powder using an e-nose based on MOS sensors. The gas chromatography-based e-nose was used to inspect the adulteration of honey with corn and rice syrup (Gan et al., 2016). For commodities like meat, aroma is the main indicator to be used in e-nose technology. This is because meat's lowered shelf-life stability made the aroma more vulnerable. An e-nose has been successfully used to detect the adulteration of various meats. Tian et al., (2019) reported the minced mutton was adulterated with pork using a PEN 2 e-nose system containing ten selective MOS gas sensors based on the odor fingerprint. Wang et al. (2019) also successfully detected the adulteration of mutton with duck meat using SPME for volatile compound extraction and PEN 3 e-nose system comprised of 10 MOS sensors. Another common application of e-nose is the detection of adulteration in spice. Banach et al. (2012) investigated the adulteration of 'saveloy' and 'sausage' spices with an admixture of 20% concentration level of adulteration with curry leaves and garlic spice powder using 38 MOS sensor-based commercial type e-nose (KAMINA, Yson GmbH).

5. DNA molecular-based technique

DNA is a unique organism, hence, DNA-based techniques can be an effective tool in food adulteration detection. Unlike physical, chemical and biochemical techniques, this DNA-based technique can provide an accurate result quantitatively and qualitatively. Therefore, the developed techniques such as multiplex-PCR, quantitative PCR (qPCR), DNA sequencing, restriction fragment length polymorphism (RFLP), randomly amplified polymorphic DNA (RAPD), and simple sequence repeats (SSR) or microsatellites methods are the potent technology which are required by countries and international trade organizations to agree especially on the standard. For instance, the DNA-based molecular technique can be used to detect adulteration, particularly for food with nearly similar physical characteristics. New tools, including high-resolution melting (HRM) PCR, droplet digital PCR (ddPCR), isothermal amplification (e.g., loop-mediated isothermal amplification, recombinase-polymerase amplification, strand-displacement amplification, heli-case-dependent amplification, and rolling-circle amplification), and next-generation sequencing (NGS) have emerged with a better performance in terms of sensitivity, specificity, speed, and multiplex.

5.1 PCR and qPCR

Detection of adulteration and identification of animal and plant species can be carried out using high sensitivity, specificity, and relatively fast PCR techniques. The simultaneous identifica-

tion of several species can be obtained through multiplex PCR assays using specific primers. Thus, it can detect and distinguish the species in the food products (Böhme et al., 2019). This technique allows the detection of a target in a complex matrix at a very low concentration through the exponential amplification of a target DNA molecule using a thermal cycler that generates up to millions of copies (Bohme 2019). Multiplex PCR assays targeting the sensitive qualitative region of mitochondrial DNA have been developed to simultaneously detect different animal species in meat because this region has a high cell copy number. For example, the adulteration of meatballs with meat from dog, rat, rabbit, and squirrel were identified down to 0.1% (Ahamad et al., 2017; Rahman et al., 2014). Nevertheless, the limitation of the PCR technique is not reliable for quantification because the copy numbers per cell change between species, individuals, and even tissues within the same individual.

The quantification measurement in food adulteration is very important to justify whether it is intentional or unintentional mixing. In this sense, qPCR is a reliable and convenient method to quantify adulterated food products. The principle of qPCR is amplification of nucleic acids, which can be monitored in real-time, and quantification by measuring the fluorescence coming from the release of double-strand-DNA-binding dye, which is measured in each PCR cycle (Böhme et al., 2019). The performance of qPCR offers high sensitivity, multiplexing, high speed, and relatively low cost compared to the conventional method. Furthermore, this technique does not require the post-PCR processing steps like in the conventional method because it is based on real-time monitoring of the increasing number of target DNA molecules. This qPCR technique has successfully detected the presence of forbidden meat species like pork in raw and processed halal food products (Karabasanavar et al., 2014; Sakalar et al., 2015). Besides that, it has been used in the seafood industry to detect fraud or mislabeling in tuna products (Liu et al., 2016). However, the effects of tissue composition and matrix components on the PCR efficiency and precision are the main limitations on the absolute or relative quantification.

5.2 DNAFoil

DNAFoil is a novel technology based on DNA-approach can be used for food adulteration detection. It is portable, completely self-administered, and can be performed on-site without expensive equipment such as PCR or any experimental work to validate the food adulteration in a very short time, as little as 30 minutes. Figure 4 illustrates the process flow and mechanism of the DNAFoil technique.

Herein, this technique can detect food adulteration by following these five main steps (Sheikha, 2019): initially, the provided barrel to break, lyses, extract, neutralize, and stabilize DNA in the food sample. Subsequently, the DNA target amplifies by transferring one drop of the extracted DNA (step 1) to the reaction tube, which is then placed in the water. The DNA target will amplify and make multiple copies using enzymes and specific primers identified. Finally, immerse the revealing strip after waiting about 30 minutes to detect any traces. The target DNA will be transferred via capillary force to the detection surface of the test strip during the final stages. The target DNA fragments will be captured in a band on the test strip. These procedures show the versatile, potent, and rapid technology of DNAFoil.

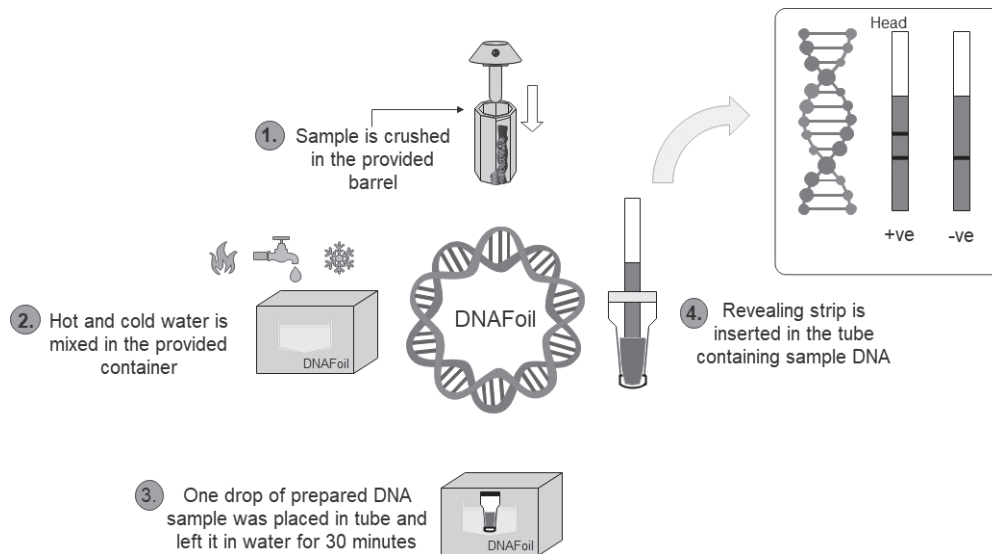


Fig. 4 Process flow and mechanism of DNAFoil technique (Sheikha, 2019)

This DNAFoil kit technology has successfully detected pork adulteration in beef at lower levels, which was reported in Meat and Livestock Australia Limited in January 2018. The DNAFoil validity has been confirmed using qPCR. Aronoff et al. (2018) demonstrated the reproducibility data to detect the vegetal material in milk products using qPCR. This finding indicates this DNAFoil kit technology is a rapid and reliable method to validate product content. It can also be used to identify the characteristics in minimal time without needing any lab equipment, technicians, or scientists.

6. Conclusion

The development of food integrity management system within food supply chain is important to create trust and brand integrity among stakeholders. This can be achieved through a few contexts by ensuring: (i) safety, quality, and authenticity of food products, (ii) reliable labelling, and (iii) effective management of provenance such as halal status, vegan, or organic product. In the sense of the first context, adulteration and authenticity is a key issue that has been gaining increasing attention by all players including industry, government, academia, and consumers in recent years.

A tremendous improvement on the analytical methodologies to investigate the food product integrity especially in detecting or analyzing the adulterants are seriously in need due to the countless food scandals have evolved. This is important to gain the consumer confidence and protecting consumer rights by guarantee the quality, safety, and authenticity of the food product as well as processing methods according to the standard operating procedures.

For this reason, this chapter reviewed the food integrity in the perspective of food science and technology on the safety, quality, and authenticity of the food product within the wider food supply chain. With an increasing crises and food industry scandals, a rapid analytical technique has been developed to carter with this issue. Herein, the available analytical methods particularly

for chemical/biochemical and DNA-molecular technique have been reviewed. This review indicates a significant enhancement of the analytical methods development to mitigate the adulteration or authenticity issues by promoting several advantages such as high efficacy, rapid, and minimal cost. Nevertheless, there still need a continuous research and development to improve the limitation of the techniques.

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