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Food authentication with novel analytical approaches: A review

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Abstract

Food Authentication is becoming an emerging field owing to rapid growth of public concerns over safety and quality of food. This article explains some new analytical techniques which are being used for assessment of food authenticity. These technologies are classified on the basis of authenticity indicators. Recently developed novel analytical techniques and their plausible solutions in authenticity problems are discussed. The article also explains the limitations and gaps which are related to handling of large experimental data. To progress the assessment of food, reference database and standards are required. A scientific evaluation of new methods featured the research prospects and arising advancements in this evolving field, while the capacity of the field for future use is illustrated in the secular manner of the research outcomes, emphasizing on geographical dissemination of research analysis and preferred forums used for distribution.

Keywords: Food; Safety; Quality; Authenticity; Analytical; Techniques; Assessment; Indicators; Evaluation; Geography; Adulteration.

1. Introduction

Food authentication is a phenomenon about compliance of food with its label instructions. It covers the food origin (geographical or generic background), production methodology (conventional or non-conventional) or processing techniques (sterilization, freezing and irradiation). The description about quality characteristics in value added products has gained much importance because of the rising fraudulent labeling that has become a social issue. Proof about originality is highly important topic regarding food safety, its quality and consumer protection (Aung and Chang, 2014). The rapid expansion of food items in market and the resulting ease in availability of food products from all over the world has enabled the consumer to take interest about food safety. The methods involved in food authentication are highly important regarding commercial and legislative concerns (Drivelos and Georgiou, 2012).

Food authenticity has now become a major issue for consumers and regulators. Sophisticated technologies in science along with modern communication ways have developed precise measurement to determine origin of food (Georgiou and Danezis, 2015). Since 20th century, the organizations involved in standardization and authentication of ingredients, have been distributed worldwide. The food items that meet standards are generally considered better in quality and generate more profit with some avidity that diverts the mind to produce adulterated food stuffs (Riedl *et al*., 2015).

Authenticity determination is criterion for safety of food materials. Thus, there is need to utilize food authentication technologies for protection of regional foods (Posudin *et al*., 2015). Food authenticity serves all the stakeholders including consumer and food industries which want to assure that their products meet legislation and labeling standards (Reinholds *et al*., 2015). Now there is a demand by regulatory authorities to update the approaches for confirmation of food authenticity, food laws and regulations. This review focuses on advances in technologies regarding food authenticity.

2. Analytical approaches

In addition to the already described views, precise and maverick analytical techniques are also required to determine certain parameters of authenticity. Normally, the authenticity or originality of food can be determined using a sufficient quantity of valid and stable biomarkers (Reinholds *et al*., 2015).

2.1 Molecular methods, Genomics-Proteomics

A wide range of analytical methods for authentication of food origin have been devised and tested. Molecular techniques for testing variation of original food stuff, is a vast molecular methodology. These novel techniques are rapidly surpassing the conventional methods. The genomics and proteomics techniques are mostly applied for species identification and analysis (Patra *et al*., 2017). Since the somatic cells have identical DNA in a given living organism, it remains unchanged when extracted from any living tissue. As DNA is highly stable in nature so it can be used to analyze excessively processed food stuffs with trace contaminants. (Kussmann and Bladeren, 2011). These genomic techniques for food fraud indication depend on specific clarification of DNA constitutes by the process of Polymerase Chain Reaction (PCR). This method involves genomics because the complete genome of food sample is studied (Ali *et al*., 2014). The proteomics involves the proteins which can behave as marker for several attributes of food stuffs so it can indicate the new peptides in sample. The main advantage of using genomics is the measurement of a trace amount of nucleotide material, while the proteomics involves identification of encoded products by DNA. These methods are highly sensitive which means that a very small amount of sample is required. After the initial discovery using reference materials, analytical methods are required for targeted identification and measurement of markers that are characterized in unknown food samples (Gallardo *et al*., 2013) These proteogenomic methods are being modified, like PCR-SSCP (PCR Single strand conformation polymorphisms), DNA-RAPD (Random amplified polymorphic), or the latest field of (PNA) Peptide Nucleic acid and fingerprinting with DNA (Sforza *et al*., 2011). The methods of genomics and proteomics techniques are used to examine fake description and labeling issues of foods, example including GMOs detection, authentication of seafood, identification between halal and kosher meat, detection of pork meat in products having description as beef, authentication of game meat, food botanical origin, authentication of species origin (Seçkin *et al*., 2017). There is another field underlying in proteogenomics is named as microbial fingerprinting. PCR DGGE (PCR Denaturing Gel Electrophoresis technique) is normally used for this type of fingerprinting. This technique is mostly applicable in cheese (Arcuri *et al*., 2013). Other food products such as milk, cocoa and organic foods are also verified by that method.

2.2 Chromatographic techniques

These methods are feasible and provide rapid separation of chemically identical compounds in food matrices (Cserháti *et al*., 2005). Food matrices consist of variety of compounds, including carbohydrate, proteins, lipids, fatty acids, amino acids, nucleic acids, organic acids, phytochemicals and other molecules, like aromatic and colorants (Ibáñez *et al*., 2013). These constitutes are diverse, having different range of size and polarities. Chromatography techniques show unique phenomenon of fingerprinting for food authentication. The analytical variation between patterns of marker compounds is principle of chromatography.

The foodstuffs have complex chemical structure and the high demand about quality and originality requires high resolution techniques like gas chromatography (GC) and liquid chromatography (LC) combined with mass spectrometry (MS), are considered best for use (Gallardo *et al*., 2012).

LC base separation is employed by targeting three main characteristics of compounds: electric charge, polarity and molecular size. It is considered feasible to detect carbohydrates, proteins, vitamins, phenolic compounds, chiral compounds, and various pigments, while gas chromatography is suitable to volatile and semi-volatile compounds (Di Stefano *et al*., 2012).

The chromatography methods are widely used in food authentication including adulteration determination in honey, milk, cheese, olive oils, mushrooms and vegetables. This phenomenon of authentication is done by comparing profiles of measure compounds with predetermined values (Cuadros-Rodríguez *et al*., 2016).

2.3 Isotopic ratios techniques

The Isotopes have same empirical formula but differ in molecular weight. Stable Isotopes are divided into two classes on the basis of atomic mass (the light and heavy isotopes). In food authentication the isotopic ratios of these classes are investigated. The isotopic ratio analysis involves various methods including Isotope Ratio Mass Spectrometry (IRMS), Multi Collector Inductively Coupled Plasma Mass Spectrometry (MC-ICP-MS) and Thermal Ionization Mass Spectrometry (TIMS) (Kelly *et al*., 2005).

The Isotopic ratios vary with environmental conditions, pedology and geographical origin of food. The H and O isotopic data of food are linked to isotopic data of water having geographical variability. The climatic relation to isotopes is usually linked to N and C, agricultural practices, geology, distance from the sea and volcanism effect on S isotopes. The analysis regarding stable isotopes have been described by AOAC methods since 1990s to determine adulteration in various food items including, honey, wine and fruit juice with inexpensive extenders like sugar syrup and water (Vinci *et al*., 2012). The difference between natural and synthetic vanillin can also be determined by isotopic ratios analysis. Recently, the multi isotopic ratio method has been used for verification of wine, orange juice, orange, tomato, olive oil, cheese, meat and dairy products. The originally cultivated products are indicated by isotopic fingerprinting techniques**.** This fingerprinting phenomenon can be coupled with other methods like GC and NMR to improve the verification ability of analysis **(**Hohmann *et al*., 2015).

2.4 Vibrational and fluorescence spectroscopy

Vibrational spectroscopy is considered the rapid and cheaper method for food quality assessment along with food authentication. Modern instrumental methods coupled with chemometric techniques have allowed the introduction of fast methods which use multivariate analysis (MVA) to near infrared and mid infrared range to determine food matrices. The samples of different phases like gas, liquid or solid, can absorb infrared radiations in infrared region and produce fingerprints at specific frequencies. The bending, stretching, and rotating phenomena in molecules, induces MIR fingerprinting (Ghidini *et al*., 2019). While NIR spectrum is produced due to overtone various combinations of frequencies. Another emerging technology, called Raman spectroscopy is relied on vibrational phenomenon which can be targeted to specific functional groups and can prove to be useful for fingerprinting of food samples. The detection limit of Raman spectroscopy is very high so it is more feasible for qualitative analysis. The analysis through Raman spectroscopy is rapid and non-destructive. SERS (Surface enhanced Raman spectroscopy) has low detection limit and it is more feasible for determination of adulterants in food (Cozzolino, 2012).

There is another non-destructive, simple, and inexpensive technique called fluorescence spectroscopy having detection limit relatively low. This technique usually detects polyaromatic hydrocarbons and heterocyclic compounds with definite molecular structure. Recently, fluorometers have been developed that provide accurate results in low cost by coupling with other sophisticated techniques (Ahad and Nissar, 2017). This technology provides more spectral elaboration with rapid and repeatable results. There is another technique known as SyF (Synchronous fluorescence) that enhances the differentiation ability of fluorescence by using excitation-emission spaces and food adulteration (Poulli *et al*., 2007).

Spectroscopic techniques are being applied in wide range of food products for detection of adulterants like melamine in milk, syrup and maltose detection in honey, vegetable oil detection in olive oil, detection of millet and wheat in grounded black pepper, colorant detection in spices and detection adulterants in meat. Authentication of olive oil, honey, wine, milk, spirits and other ingredients have been performed by utilizing this technology (Ruoff *et al*., 2006).

2.5 Elemental methodologies

Food authentication can also be assessed by elemental profiling techniques. The elemental profiling consists of macro-elements, micro-elements and rare earth elements. Soil is the main source of these elements for plants but there are various factors that affect the concentrations of these elements like fertilization application, botanical origin, pollution, harvesting and production period (Fragni *et al*., 2018). The changes in concentration are however smaller than those observed between geographical regions and production regions. Geographical origin of food can be effectively determined by using rare earth elements as the fingerprints of these elements directly indicated the geology without any significant influence by other factors, such as production year and agricultural practices. The elemental profile of food commodity indicates the mineral composition of fodder and other feed. The other factors that determine elemental content are drinking water, soil composition and pollution and these rely on geographical origin (Furia *et al*., 2011).

There are various analytical techniques that measure elemental fingerprinting of food. In past, atomic absorption spectroscopy has been used. The modern methods utilize ICP-MS (Inductively Coupled Plasma Mass Spectroscopy) and ICP-AES (Inductively Couple Plasma Atomic Emission Spectroscopy) for multi element analysis (Vogl, 2013). The elemental fingerprinting techniques use to discriminate conventional foods from organic foods. This technology is appropriate for determining authenticity of honey, spices, olive oil, coffee, wine, vegetables and fruits (Laursen *et al*., 2014).

2.6 Nuclear magnetic resonance (NMR)

NMR is an efficient technique to get high throughput spectral and structural information about a series of compounds. It enables highly precise measurement of complex food matrices. The quantity of any component in a food mixture can be analyzed with few sample preparations. The main disadvantage of NMR considered in past, was its sensitivity, but continuous improvement in sensitivity has been done in modern hardware. Now NMR utilizes collection of metabolic profiles that can help in food authentication (Hatzakis, 2018). SNIF-NMR, which is abbreviated as (Site Specific Natural Isotopic Fractionation) is a modification of NMR and this is used for robust fingerprinting of molecules. The geographical origin of various foods like wine can be assessed by this technique. NMR can be utilized to detect adulterants in many food items, such as detection of anthocyanins in red wine, synthetic food grade flavors in natural flavors, corn syrup detection in maple syrup (Longobardi *et al*., 2013).

2.7 Sensory analysis in food authentication

Nowadays, there is high demand of food products with extraordinary organoleptic characteristics. Sensory evaluation of food is becoming more important for food stakeholders. To carry out sensory analysis, a well-trained team of sensory assessors is required. Sensory test panel uses a series of techniques to assess the organoleptic behavior of food products (Elortondo *et al*., 2007). Color, flavor, texture and aroma are the primary attributes of food quality or authenticity. The sensory penal requires extensive training, proper replication and statistical analysis of measured observations. The sensory assessment may differ among panelists because of variation in sensitivities and preferences so there is a need of reasonable perception and commitment (Śliwińska *et al*., 2014).

It is necessary to have standardization of these methods and this can only be achieved through instrumental techniques. These methods are rapid and can recognize sensory perceptions in same manner as expert panelists. Now, more attention is given to perception sensors that can test food quality. These techniques are GC-O (Gas Chromatography-Olfactometry, e-nose (electronic nose), e-tongue and e-eye (Wojnowski *et al*., 2017). The detection of volatiles produced on headspace can be detected by using e-nose. The food sample is volatilized and detection system containing sensors examines these volatile compounds. The physical change is measured when compound absorbed on the surface of the sensor. The readout system convert received signals into digital language. The sensors are specific to the volatile compounds. The results are then compared with statistical standards. The result from instrumental techniques can also be compared with those of manual sensing. The major advantages of instrumental methods include minimal sample preparations, simple and easy procedure, fast and inexpensive (Kandpal and Cho, 2014).

2.8 Mass spectrometry

These methodologies comprise of most efficient ways to determine food authenticity including, DART-MS (Direct Analysis Real Time Mass Spectroscopy) and PTR-MS (Proton Transfer Reaction Mass Spectroscopy). PTR-MS is a quantitative technique to measure volatile compounds by applying chemical ionization (soft) method for ionization of molecules (Gerbig *et al*., 2017). The volatile compounds react with charged ions that give protons to volatile compounds and yield protonated analyte. The electric field then takes these ions into reaction chamber and collision induced ion dissociation occur. As a result fingerprint of sample is obtained. PTR-MS provides absolute concentrations of volatile samples with rapid detection (Luykx and VanRuth, 2008).

DART-MS is also known as ambient mass spectrometry technique and it is performed in an open environment. It is greatly simplified and fast method of detection. In this method, a gas with high ionization potential is used in which electrical potential is applied to form excited atoms or molecules and these molecules remove lower mass atoms from the sample surface. DART-MS can also be used in analysis with high salt tolerance. Mass spectrometry techniques are best to detect food authentication and determination of fake or incorrect labeling. These methods can also be used to determine the botanical origin of foods (Li *et al*., 2015).

2.9 Immunological methods:

These methods rely on affinity of antibodies to react with respective antigens. In Past these methods were used to immunology but now are widely used in food fraud indication. The analytical tools or immunoassays are efficient ways to identify food standards because of rapid, highly specific and inexpensive features. Food industries prepare antibodies antagonistic to antigens as capturing agents. The development of crucial antibodies is crucial and highly sensitive step (Gajewski and Hsieh, 2009).

Enzyme linked immune sorbent assay, best known as ELISA, is a useful immunological methods for food authentication. It is widely used in authentication of meat and milk products. It can also detect the genetically modified microbes and radiation treated products (Asensio *et al*., 2008). ELISA is famous for determination of bovine growth chemicals and malamine in milk and identification of pork meat in ground beef (Santaclara, 2014).

3. Conclusions

Food authentication comprises of number of disciplines including, biology, mathematics, statistics, bioinformatics and agricultural sciences. So the analytical scientists lead for the development of sophisticated techniques for food authentication on the basis of their respective field of interest. This review summarizes brief study about analytical techniques, information needs and latest scientific evaluation of the field. The widest utilization of methods is the verification of food botanical origin. Mass spectrometry is becoming most efficient method in food authentication because of high throughput and sensitivity. Multi sample capability is now becoming an essential requirement because a vast size of data for analysis is being generated and handling or managing high volumes of information is a crucial process.

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Table 2.1 Analytical techniques for authentication of different food

Food	Purpose	Analytical techniques
Meat and fish	Pork adulteration in beef	FTIR
	Beef production system	NMR
	Beef geographical origin	NMR
	Farming system of fish	NMR
	Animal species identification	ELISA
Dairy products	Milk adulteration	FTIR/NIR
	Geographical origin of cheese	NMR
	Milk species	ELISA
Honey	Geographical origin	NMR/NIR/GC-GC/Raman
	Botanical origin	NIR/FTIR/GC-MS/NMR
	Adulteration	FTIR/NIR/Raman

(Abbas *et al*., 2017)