

Food Forensics Analysis of Food Products from Forensic Science

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ABSTRACT

Food forensics is an interdisciplinary field that help in the analysis of food products with the application of forensic science that helps us to meet the standards of food products and ensuring the safety, authenticity of various food products. Forensic Sciences use different scientific methods to detect contamination, adulteration and fraud in food items. To conduct the analysis various methods are used in this field such as mass spectrometry, DNA based techniques, chromatography, PCR, HPLC, sensory evaluation, Nuclear magnetic resonance spectroscopy. This instrument helps in the analysis of various toxins present in the food product and even pathogen detection. This sophisticated technique helps us to analyze the food sample properly so that it can be used in our day to day lives for the consumption. Authenticity research funded by the Food Standards Agency (FSA) is driven by the need to verify compliance with food standards and labelling legislation and to detect food fraud. Method development has therefore been designed to verify legal requirements relating to the names given to food, the name and quantitative declarations of food processing or treatments, and claims of production and geographic origin. A major focus of food forensics is detecting food fraud, which includes misinterpretation, ingredient tampering and counterfeit products. For example DNA barcoding can confirm the identity of meat or seafood, preventing fraudulent species substitution. Regulatory bodies, including the FDA, EFSA AND WHO, Depend on forensic food analysis to uphold food safety regulations and prevent the distribution of unsafe products. With the increasing pervasiveness of food fraud and safety concerns, food forensics has become essential in ensuring public health and protecting market integrity. The advancements in forensic science will further enhance the effectiveness and precision of food analysis reinforcing its importance in the global food industry.

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INTRODUCTION

In an age where global food supply chains grow ever more complex, the assurance of food authenticity, safety, and integrity has become a matter not merely of commerce, but of public health, ethics, and trust. **Food forensics** emerges as a multidisciplinary science devoted to the *detection, investigation, and prevention* of food-related crimes, frauds, and contaminations.

Rooted in the principles of analytical chemistry, molecular biology, microbiology, and law, food forensics seeks to

answer pressing questions such as: *Is this food what it claims to be? Has it been tampered with or adulterated? Can we trace its true origin?* From unearthing counterfeit extra virgin olive oil to tracing the geographical source of a protected wine, the field applies rigorous scientific techniques to uphold truth and safety within the food system.

Food fraud—defined as the intentional misrepresentation of food for economic gain—costs the global economy billions annually and often places consumers at serious risk. Scandals such as the 2008 melamine-tainted milk crisis in

China or the 2013 European horse meat scandal illustrate the need for forensic scrutiny. Beyond fraud, food forensics also deals with unintentional contamination, allergens, microbial threats, and even criminal acts like deliberate poisoning or bioterrorism.

By integrating classical investigative techniques with cutting-edge technologies—such as DNA barcoding, isotope analysis, and mass spectrometry—food forensics provides a robust framework to uncover hidden truths in what we consume. It serves not only the interests of public health and consumer rights, but also reinforces a broader ethical imperative: to preserve honesty, transparency, and justice in one of humanity's most essential domains—our daily bread.

Food forensics uses scientific methods to authenticate and trace foodstuffs. Forensic Chemistry employs chemical methodologies to assist investigative organizations and law enforcement. Food forensics is an academic discipline that examines explicitly and investigates any unlawful activities associated with food items or their use within a particular region. Further, adulteration refers to the intentional contamination of food with substandard or inexpensive, inedible, or poisonous chemicals, resulting in long-term poisoning, illnesses, and potentially even deaths. Food forensic investigations have gained little popularity, particularly in India, where the Food and Drug Administration (FDA) routinely conducts most cases. A study assessed if existing methods should have been modified to address vendors' criminal intentions and regulations regarding local commodities' presence in households throughout India.

Food forensics is a proven process that applies scientific techniques to detect and identify food pollutants, hence ensuring the safety of the worldwide food supply. The process entails utilizing analytical methods to verify the authenticity and traceability of food products. Food contamination can occur through chemicals, microorganisms, or foreign objects. Chemical contamination can occur either through direct exposure or indirect means, such as the presence of dangerous chemicals, pesticides, or additives. Indirect contamination arises because of the application of pesticides for crop protection. Biological contamination refers to the presence of microorganisms that result in alterations in sensory perception. Spoilage organisms that are the primary sources are observed in air, insects, water, dust, factory workers, employees, and stray animals.

Role of FSSAI In food forensics

The Food Safety and Standards Authority of India (FSSAI) is crucial in the realm of food forensics within India, even though its main function is to regulate food safety. Regarding food forensics, the role of FSSAI encompasses a mix of regulatory, scientific, and enforcement duties that work to guarantee the authenticity and safety of food items.

Here's a detailed look at its responsibilities:

1. Identification of Food Adulteration and Contamination

- FSSAI is tasked with detecting and controlling food adulteration, which is a significant focus of food forensics.
- It establishes standards for allowable levels of additives, contaminants, and pesticide residues.
- Scientific techniques are employed to identify biological, chemical, and physical adulterants in food samples.

2. Forensic Analysis and Laboratories

- FSSAI operates a network of National Food Laboratories (NFLs) and Notified Food Testing Laboratories that are equipped with forensic and analytical capabilities.
- These laboratories carry out:
 - DNA analysis to confirm food authenticity (such as verifying the source of meat)
 - Mass spectrometry for identifying chemical adulterants
 - Spectrophotometry and chromatography to detect contaminants

3. Legal Enforcement and Support for Prosecution

- Food forensics aids FSSAI in providing evidence for legal proceedings against offenders in accordance with the Food Safety and Standards Act, 2006.
- FSSAI works alongside law enforcement and legal entities to prosecute cases of food fraud and adulteration with the help of forensic reports.

4. Research and Development

- FSSAI dedicates resources to R&D to create innovative methods in food forensics, including systems for traceability and AI-driven tools for anomaly detection in food products.

5. Partnerships

- FSSAI collaborates with international organizations (such as Codex Alimentarius, WHO, etc.) and academic institutions to enhance forensic capabilities.
- Additionally, it engages with customs and trade authorities to prevent the import and export of adulterated or improperly labeled food.

FOOD SAFETY FORENSICS

Food Safety Forensics refers to the approach of applying food safety principles, detection techniques, and processes to investigate crimes or to confirm and document foodborne illnesses or contamination affecting both humans and pets. This methodology focuses specifically on foodborne microorganism poisoning and provides a structured approach to determine the causes of food poisoning and the

factors involved, outlining a series of investigative steps for tracking and tracing, along with relevant technologies and detection methodologies. A pertinent example is the detection and tracing of pathogenic organisms in products like melons in Colorado that led to fatalities among consumers. Forensics is typically defined as the use of scientific methods to resolve criminal cases. Food Science Forensics is the application of food science principles and procedures to address crimes or to confirm and document cases of food poisoning due to adulteration affecting humans and pets. This approach is not limited to foodborne microorganism poisoning. It involves a systematic methodology to identify the cases of food poisoning and related factors, specifying a sequential process of "tracking and tracing" investigative steps, technologies, and detection tools. A recent example includes the identification and tracing of arsenic contamination in rice products from their origin.

Comparison of Food Forensics with Other Forensic Disciplines Food Safety Forensics (FSF) is distinct from the other nine forensic disciplines in several significant ways:

- Food, similar to pharmaceutical substances, is consumed by individuals.
- Food can be a matter of life and death.
- Food items are perishable and have a limited shelf life - their "fingerprints" can disappear quickly.
- Food Safety Forensics integrates various methodologies from other forensic fields.
- Food Safety Forensics methodology must encompass the complete "farm-to-fork" supply chain.

A Brief History Of Food Forensics

Antiquity: Ethical and Legal Origins Long before the development of laboratories and spectrometers, ancient cultures acknowledged the significance of honest food commerce.

Ancient Egypt and Mesopotamia: Food regulations were established in early legal systems. The Code of Hammurabi (around 1754 BCE) included measures against food fraud, such as dishonest weights and measures for grain and beer.

Ancient Greece and Rome: Thinkers like Galen and Hippocrates regarded diet as essential for health, and Roman law (under emperors like Trajan) began overseeing food markets. Olive oil, wine, and bread were frequently monitored by public officials.

Early Christianity and Islam: Both faiths strongly denounced food adulteration as a transgression against justice and the collective welfare, with Islamic law (Sharia) mandating the purity of halal foods through stringent regulations.

Middle Ages to Enlightenment: Observation and Distrust During the medieval period, city officials started inspecting food in expanding urban areas. Guild systems across Europe regulated the production of bread, wine, and meat to prevent deception.

In England, the Assize of Bread and Ale (13th century) set standards for weight and quality, forming early foundations for food regulation. However, scientific verification was absent; detection often depended on taste, smell, or basic visual indicators.

19th Century: Emergence of Analytical Chemistry and Consumer Safeguards The industrial revolution brought forth processed foods, alongside widespread adulteration.

Frederick Accum (Germany/England, early 1800s): One of the earliest chemists to alert the public about food fraud. His publication

"A Treatise on Adulterations of Food" (1820) revealed practices such as adding copper to pickles for color or alum to bread.

Arthur Hill Hassall (mid-1800s): A trailblazer in microscopic analysis of food in Britain. His revelations sparked public outrage and prompted legal changes. These initiatives contributed to the formation of early Food and Drug Acts, including:

The UK's Adulteration of Food and Drink Act (1860). The Pure Food and Drug Act (1906) in the United States, influenced in part by Upton Sinclair's "The Jungle."

20th Century: Science Assumes Control As scientific progress continued, food forensics became more advanced. The development of chromatography, spectroscopy, and DNA analysis facilitated precise identification of adulterants, toxins, and biological indicators. Significant food scandals (e.g., tainted baby formula, prohibited dyes) led nations to create regulatory bodies: FDA (United States).

EFSA (European Union). FSSAI (India), amongst others. **21st Century: Globalization, Genomics, and Digital Traceability** Currently, food forensics operates as a high-tech, interdisciplinary field. DNA barcoding can identify species in meat, fish, and herbal products—revealing mislabeling and safeguarding endangered species. Isotope ratio analysis assists in tracing geographic origins (e.g., is that "Champagne" genuinely from France?).

Blockchain and AI are being investigated for enhancing supply chain transparency. Following scandals such as the 2013 Horse Meat Scandal and counterfeit honey imports, governments have allocated resources to advanced food crime units.

REVIEW LITERATURE

Advanced detection tools in food fraud: A systematic review for holistic and rational detection method based on research and patents

Food fraud and adulteration have become urgent global issues, carrying serious consequences for public health, economic stability, and consumer confidence. Numerous high-profile cases highlight the gravity of this problem. For instance, the 2008 scandal involving melamine-contaminated milk in China caused over 300,000 illnesses, six infant fatalities, and numerous hospital admissions, showcasing the extreme dangers associated with manipulating food products to meet supposed nutritional requirements (El Benni et al., 2019; Liang et al., 2020). In a similar vein, the 2013 European horse meat scandal and the 2009 Italian olive oil fraud exposed systemic vulnerabilities in food labeling and supply chain transparency (Ribani et al., 2018). Public health continues to be a significant issue, as contaminated food can result in both acute and chronic health problems. Repeated dioxin contamination incidents in the EU, along with widespread adulteration of spices, dairy items, and meats, underline persistent hazards (Focker et al., 2022; Singh et al., 2021). Additionally, the presence of synthetic additives in products like pickles, yogurt, and cheese, including artificial colors, plant oils, and gelatin, further heightens safety concerns (Cavdaroglu & Ozen, 2023; Usman et al., 2022). From an economic perspective, food fraud is estimated to cost the global economy around \$40 billion each year, driven by the desire to dilute high-value goods with lower quality substitutes (Zhang et al., 2020). This economic effect is made worse by the erosion of consumer trust and legal repercussions, as exemplified by the widely reported Subway tuna controversy (Robinson, 2023). On the technological front, identifying food adulteration remains difficult due to the constantly evolving tactics employed by fraudsters. Conventional techniques such as gas chromatography-mass spectrometry (GC-MS), liquid chromatography (HPLC, LC-MS), and electrophoresis are being augmented by cutting-edge molecular and machine learning technologies (He et al., 2021; Lin et al., 2023). New tools like smart electronic noses and CRISPR-based diagnostics are being increasingly incorporated into food monitoring frameworks (Spink et al., 2019). In summary, the existing literature indicates that food fraud is a complex threat necessitating coordinated efforts on a global scale. Although progress has been made in detection technologies, the financial motivations behind fraudulent activities, alongside regulatory gaps and intricate international supply chains, continue to pose challenges for prevention and enforcement.

FOOD-PRODUCING ANIMALS AS IMPORTANT RESERVOIRS OR CARRIERS

The relationship between foodborne pathogens and animal agriculture has been the subject of extensive research, particularly in light of the significant public health

implications. Painter et al. reported that although plant-derived foods are responsible for the majority of foodborne illness cases, animal-derived commodities are more commonly associated with fatalities. This disparity highlights the severity of pathogens typically linked to animal products, many of which can cause life-threatening conditions in humans.

Food-producing animals serve as natural reservoirs for numerous foodborne pathogens. These microorganisms often reside in or on the animals without causing overt disease, thereby complicating detection and prevention efforts. For example, cattle are known asymptomatic carriers of *Escherichia coli* O157:H7, a pathogen capable of causing severe illness in humans, particularly when contaminated meat is consumed raw or undercooked. Similar risks are associated with other animal products: *Yersinia enterocolitica* and *Trichinella* spp. are commonly found in pork, while poultry is frequently contaminated with *Salmonella* spp. and *Campylobacter*.

The transmission of these pathogens is often exacerbated by poor hygiene practices during slaughter and processing. Pathogens such as *Listeria monocytogenes*, *Clostridium botulinum*, and *Bacillus cereus*—which are prevalent in farm environments—can be transferred to animal carcasses and subsequently enter the food supply. Additionally, raw milk from healthy cows may become contaminated due to contact with dirty udders, bedding, soil, or fecal matter, all of which are common vectors for microbial transmission.

Fecal contamination, in particular, is a significant source of foodborne pathogens. Enteric bacteria such as *Salmonella* and virulent strains of *E. coli*, both members of the family *Enterobacteriaceae*, are abundant in animal feces and can contaminate meat and other food products during processing. This has led to the recognition of animal farms as primary sources of foodborne outbreaks. However, cross-contamination can also occur in produce farms due to the presence of feces from wild or domestic animals, further complicating the food safety landscape.

A notable example of non-meat-related contamination is the 1996 outbreak of *E. coli* O157:H7 linked to unpasteurized Odwalla apple juice. This outbreak resulted in 65 reported illnesses and one fatality—a 16-month-old girl. Investigations suggested that apples used in the juice may have been contaminated with fecal matter from either deer or cattle in the orchard. This case underscored the importance of implementing kill steps such as pasteurization even for foods previously considered low-risk, such as acidic fruit juices.

Beyond acute illness outbreaks, there is increasing concern over the role of animal agriculture in the emergence and spread of antibiotic-resistant pathogens. The routine use of antibiotics in livestock has been hypothesized to create selective pressure that facilitates the transfer of resistance genes among commensal and pathogenic microbes. For instance, multidrug-resistant *Salmonella enterica* serovar

Typhimurium DT104 has been implicated in human infections, raising concerns about the direct link between farm practices and public health.

However, findings from whole-genome sequencing (WGS) studies challenge the assumption of widespread transmission between human and animal reservoirs. Mather et al. analyzed national collections of *S. Typhimurium* DT104 isolates and found limited overlap between antibiotic-resistant strains in animals and those in humans, suggesting largely separate evolutionary pathways. Nonetheless, the possibility of cross-transmission in specific outbreaks cannot be excluded, emphasizing the need for robust surveillance and source-tracking capabilities.

In response to these challenges, regulatory bodies have advocated for the routine monitoring of fecal bacterial indicators in both raw materials and finished food products. While such indicators help assess hygiene and contamination risk, they often fall short in identifying the precise origin of pathogens. As genomic and molecular tracing technologies advance, there is potential for more targeted interventions aimed at reducing foodborne risks originating from animal agriculture.

Azodye-based colorimetric sensor array for identification of biogenic amines: Food forensics by portable RGB-based signal readout

Foodborne illnesses remain a significant global health concern, with the World Health Organization (WHO) estimating 600 million cases and 420,000 deaths annually due to contaminated food. Diarrheal diseases, largely caused by unsafe food, contribute over 550 million cases per year. Ensuring food safety across all stages of the food supply chain is, therefore, essential.

Among the major chemical hazards in food are **biogenic amines (BAs)** such as histamine, putrescine, cadaverine, spermine, and spermidine. Although these compounds play vital physiological roles, excessive concentrations (above 900–1000 mg/kg) pose serious health risks, including toxicity and carcinogenicity. Consequently, their **rapid and accurate detection** has become a priority in food quality control and forensics.

Conventional methods for detecting BAs include **chromatography**, **electrochemistry**, and **capillary electrophoresis**. While effective, these approaches are limited by laborious sample preparation, high costs, and lack of portability, making them unsuitable for on-site applications. Alternative techniques have emerged, such as:

- **Thin-layer chromatography** (Andrea et al.) for wine,
- **High-performance liquid chromatography (HPLC)** (Salazar et al.) for animal tissue,
- **Electrochemical sensors** based on nanomaterials

(Jhah et al.),

- **Chemoreceptive sensors** using charge transfer complexes (Chen et al.).

However, challenges such as **aqueous insolubility** and limited selectivity continue to hinder real-world utility.

Recent focus has shifted to **colorimetric sensor arrays (CSAs)** due to their visual simplicity, environmental resilience, and ability to detect a wide range of analytes. Unlike traditional "lock- and-key" sensors, array-based systems employ **cross-reactive elements** to generate distinctive "fingerprint" patterns for each analyte, analyzed using **multivariate statistical tools** such as **PCA** and **LDA**.

In this context, Singh et al. developed a **pattern-based CSA** for discriminating structurally similar biogenic amines through high-throughput screening. Inspired by these advancements, recent studies have further refined **portable, solid-state colorimetric sensor arrays** based on metal-azodye complexes, showing strong potential for practical application in food safety monitoring.

Microsatellite and DNA-barcode regions typing combined with High Resolution Melting (HRM) analysis for food forensic uses: A case study on lentils (*Lens culinaris*)

Distinguishing between plant species and their varieties is essential for agriculture, industry, and consumers. This becomes even more vital in the case of processed foods, where it is often difficult to differentiate species based on their physical characteristics; therefore, creating a reliable and robust discrimination system is critical. A common issue in grain legumes is the blending of high-quality seeds from well-known elite varieties with those of lower quality (and price) or seeds from other similar-looking species. These inferior seeds are often of lesser quality and may even pose health risks to consumers. As a result, ensuring food authenticity and certification is a prominent concern within the field of food analysis today. Specifically, lentils frequently become mixed with *Vicia* spp. This contamination with common vetch often arises because local lentil populations are grown in less-than-ideal conditions, and farmers do not pay close attention to the genetic consistency of their crops or the potential for contamination with other species. DNA-based methods have garnered significant attention for species discrimination, particularly with advancements like High Resolution Melting analysis (HRM), which assesses the temperature-dependent transition of double-stranded DNA to single-stranded DNA (Reed & Wittwer, 2004). Recent developments in this technique have resulted in enhanced resolution and precision of the instruments, as well as the innovation of saturating DNA dyes, making HRM applicable for genotyping (SNP, SSR markers) (Ganopoulos et al., 2011b, Mader et al., 2008, Wu et al., 2008) and for quantifying adulterants (Ganopoulos et al.,

2011a, Mader et al., 2011). A novel approach for identifying plant species has been introduced recently, which is based on DNA (Hollingsworth et al., 2009, Kress et al., 2005). This technique is referred to as DNA barcoding and utilizes short sequences of chloroplast or nuclear DNA (Hollingsworth et al., 2009). Plant DNA barcoding has been utilized in studies of species identification, evolution, forensics, and in recognizing species in processed plant commodities, including legumes. Recently, our team has integrated plant DNA barcoding with HRM analysis to create the Bar- HRM method, which facilitates the authentication of plant species and products with Protected Designation of Origin (PDO) status and allows for the quantification of adulterants in consumer products (Ganopoulos et al., in press). Lentils play a crucial role in human nutrition due to their high protein content (approximately 25%), substantial levels of lysine and leucine, and high dietary fiber content (87%), while being low in fat (Urbano et al., 2007). The well-known Greek lentil variety “Eglouvi,” cultivated extensively on the island of Lefkada (Western Greece), holds a strong market position and potential, as it is set to receive Protected Designation of Origin (PDO) EU status. This variety currently commands high prices (10 euros per kg), which provides a significant income for local farmers. In this study, we examined ten major Greek lentil varieties, including “Eglouvi.” By utilizing SSR markers in combination with HRM in this research, we successfully differentiated the ten lentil varieties and identified any mixtures of “Eglouvi” with other varieties. Additionally, the development and implementation of Bar-HRM targeting the chloroplast *rpoc1* region effectively discriminated between *Vicia sativa* and *Lens culinaris* species, as well as detected a contamination ratio as low as 1:100 of *V. sativa* within *L. culinaris* commercial products.

The DNA fingerprint in food forensics: the Basmati rice case

Due to recent controversies surrounding food safety, such as the presence of melamine in dairy and horse meat being sold as beef, the issue of food fraud is receiving more scrutiny. Advances in laboratory techniques are offering means to identify adulterations, with DNA analysis becoming increasingly significant. There are two main approaches in DNA analysis for food safety: (a) DNA barcoding, which is used to identify various species of plants, animals, and microorganisms, such as detecting horse meat in beef products, and (b) DNA fingerprinting, which helps distinguish between animal breeds and plant types, exemplified by the identification of inexpensive long grain rice mixed with higher-value Basmati. DNA fingerprinting relies on variations in allele sequences that

are highly changeable. These variations include microsatellites (often referred to as simple sequence repeats, SSRs, or short tandem repeats, STRs) and single nucleotide polymorphisms, SNPs, used in the second and third generations of fingerprinting, respectively. Initially developed for identifying human individuals, DNA fingerprinting has transformed forensics and enhanced efforts against crime. Testing for the authenticity of Basmati rice is a significant instance of its use in food forensics. Over recent decades, Basmati has become the preferred rice variety in the UK, partly due to the substantial population with roots from the Indian subcontinent. However, instances of adulteration with less expensive long grain rice have been common, which is sold at less than half the cost of Basmati. As a result, the British Retail Consortium (BRC), Rice Association, and British Rice Millers Association established their Code of Practice on Basmati Rice in 2005, designating 15 rice varieties as genuine. The cultivation of Basmati rice is geographically restricted to seven states in Northern India and the Punjab region of Pakistan. Article 6.1 of the Code of Practice identifies DNA fingerprinting as the standard technique to distinguish authentic Basmati from non-Basmati mixtures. This method, developed by Blich in 2000, has demonstrated its reliability and effectiveness through practical applications and numerous inter-laboratory ring trials and proficiency assessments. The Code of Practice has substantially enhanced the quality of Basmati in the European market. Between 2006 and 2010, 13 out of 54 Basmati products were found to be adulterated in various consumer tests conducted in Germany and France; however, in two recent German studies, only one out of 36 products failed to comply with the Code of Practice. Since 2005, 26 new Basmati varieties have been registered in India and Pakistan, many of which are now cultivated on a large commercial scale. As a result, the Code of Practice was revised in 2017 to now incorporate all 41 cultivars that have been released as Basmati in their countries of origin. In forensic science, criminals can only be identified based on the DNA traces left behind at a crime scene if their DNA profiles are recorded in the police database. Likewise, rice varieties can only be recognized if their DNA fingerprint is documented. Therefore, the DNA fingerprinting method originally established by the UK Food Standards Agency (FSA) needed to be modified, and the Rice Association supplied reference materials of all new varieties from official sources to our laboratory for fingerprint determination. This report presents the findings of this study, which facilitates Basmati authenticity testing in accordance with the new Code of Practice and offers additional information regarding the breeding history of Basmati varieties and their lineage from traditional ancestors.

METHODOLOGY

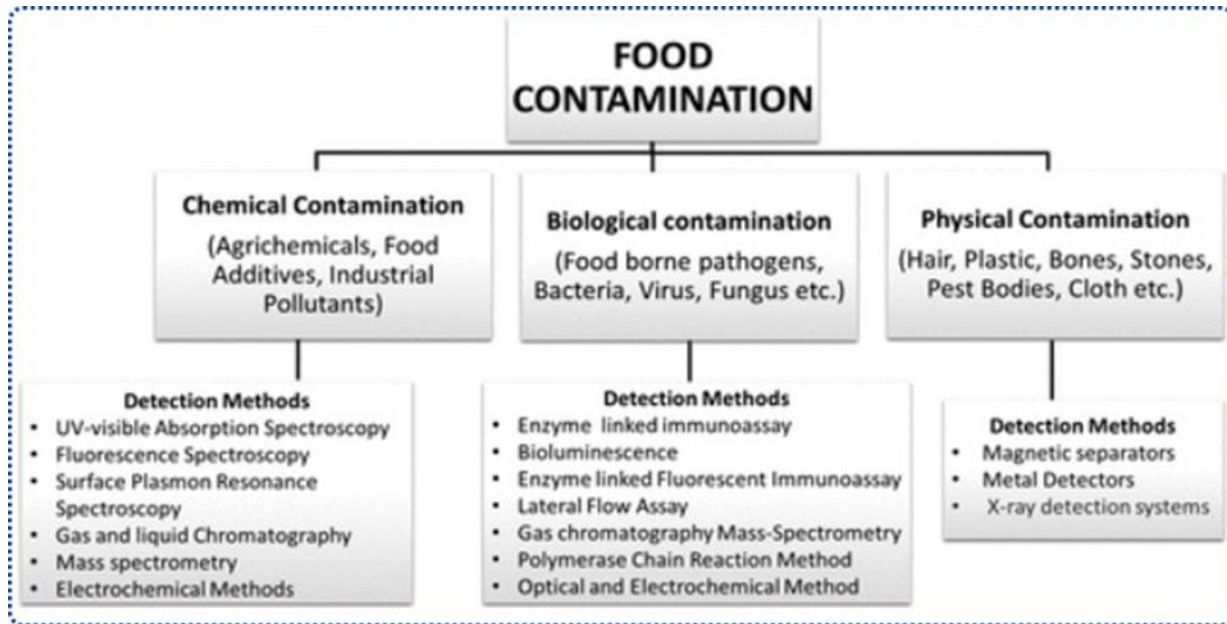


Table 1. Selected examples of misdescription of foods

Substitution of one ingredient by a similar but cheaper one	<ul style="list-style-type: none"> • Using whiting or pollack in place of cod • Substituting bonito for tuna or sea trout for salmon • Labelling cheaper varieties of potatoes as 'King Edward' variety
Extending or adulterating food with a cheaper or base material	<ul style="list-style-type: none"> • Adding water to increase weight of chicken breasts • Mixing long-grain rice with Basmati rice • Mixing cow's milk with buffalo milk before production of buffalo mozzarella cheese • Adding common wheat to durum wheat pasta labelled 100% durum wheat • Extracting soluble coffee from beans mixed with skins and husks • Adding cheaper vegetable oils to named higher value vegetable oils • Adding water, sugar, acids and colouring to fruit juices
Presence of undeclared ingredients	<ul style="list-style-type: none"> • Offal in processed meat products • Donkey or horse meat in salami • Mechanically separated meat (MSM) in processed meat products
Extending or adulterating food to increase value	<ul style="list-style-type: none"> • Adding mandarin or tangerine juice to orange juice to improve colour of juice • Adding glycerol to wine to improve body
Non-declaration or false declaration of processes	<ul style="list-style-type: none"> • Labelling poultry as fresh even though it has been previously frozen • Failure to declare that food has been irradiated • Failure to declare that juice has been prepared from concentrate
Over-declaration of a quantitative ingredient	<ul style="list-style-type: none"> • Including hydrolysed protein as part of the meat content
False claims regarding geographical or production origin	<ul style="list-style-type: none"> • Labelling South American beef as British beef • Declaring farmed fish as 'wild' • Labelling conventionally produced food as organic • Claiming that extra virgin olive oil is from a particular geographical region

1.2 USE OF STABLE ISOTOPE ANALYSIS

Hydrogen, carbon, nitrogen, and oxygen are the four primary elements found in living organisms. Each of these elements has two stable isotopes ($2H/1H$, $13C/12C$, $15N/14N$, and $18O/16O$), whose ratios can be useful in authenticity studies. For instance, the heavier isotopes of hydrogen and oxygen become more concentrated during evaporation from warm tropical and subtropical waters, and this isotopic difference, when rainfall occurs initially, can help identify groundwater originating from various geographical and climatic areas.

One of the initial applications of stable isotope analysis in

food forensics involved identifying the presence of added sugar and water in orange juice, which was also used for apple juice. The presence of added sugar can be identified if it originates from cane or maize (C4 plants) since citrus and apple are categorized as C3 plants. This differentiation is lost if the added sugar is not identifiable. is derived from beet, a type of C3 plant. Nonetheless, it is feasible to identify the presence of the added beet sugar by analyzing the $2H/1H$ ratio of ethanol produced from the sugars through nuclear magnetic resonance (NMR) (Martin, Hanote, Lees, & Martin, 1996

Food	Application
Apple juice	Detection of adulteration with cane sugar or fructose/ glucose syrups from maize (MAFF Food Surveillance Information sheet 197, 2000)
Maize oil	Detection of adulteration with cheaper oils, especially rapeseed (canola) oil (FSA Food Surveillance Information Sheet 24/02, 2002)
Palm sugar	Detection of adulteration with cane sugar (Kelly, 2009)
Beef	Verification of the origin of Brazilian, UK and Irish beef (Heaton, Kelly, Hoogewerff & Woolfe, 2008)
'Corn-fed' chicken	Verification that chickens have been fed a diet of at least 50% maize during their rearing period (Rhodes <i>et al.</i> , 2010)

One example is palm sugar, which commands a premium price and is frequently mixed with refined cane sugar. Palm is classified as a C3 plant, in contrast to sugarcane, which is a C4 plant. The presence of adulteration is identified through a technique derived from the AOAC protocol for measuring C4 sugars in honey (White & Doner, 1978) by assessing the ratio of the two carbon isotopes in the palm sugar against that of the protein extracted from the same sugar. Since the protein originates exclusively from the crude palm sugar utilized, it serves as an internal control, thereby removing the ambiguity associated with the natural variation of the 13/12C ratio in the palm sugar (Kelly, 2009).

METHODS BASED ON DNA ANALYSIS

Official food control laboratories in the UK (public analysts) generally are staffed with analytical chemists, and hence the uptake of expensive and complex molecular biology methods used for DNA analysis had been slow up to 2006. In order to encourage these laboratories to utilize DNA-based authenticity tests, the FSA put in place a technology transfer initiative, which required adapting DNA authenticity methods to a single, simplified platform for DNA detection. The platform chosen was a commercially available microfluidic lab-on-a-chip system. This permits up to 12 post-PCR samples to undergo capillary electrophoretic separation

Application	Reference
Detection/identification of 'common' meat species	Hird, Kaye, Conyers & Chisholm, in preparation
Detection/identification of 'exotic' meats	Hird <i>et al.</i> , in preparation
Detection/identification of bushmeat	Ogden & McEwing, 2008
Detection/identification of white fish species	Dooley <i>et al.</i> , 2005
Identification of salmon or tuna species in canned products	Dooley, Garrett, Sage, Clark & Brown, 2010
Identification of Basmati rice varieties and non-Basmati rice	Steele, Ogden, McEwing, Briggs, & Gorham, 2008
Detection of mandarin juices in orange juice	Scott & Knight, 2009
Detection of common wheat in durum wheat pasta	Wiseman & Burns, 2008
Identification of fruit used to make fruit pulps/purees	Clarke, Dooley, Garrett, & Brown, 2008

Transfer of an established DNA method to the lab-on-a-chip platform is not altogether straightforward, and usually requires modification or even redevelopment of the method. An example of this is the assay to authenticate Basmati rice. The original method (Bligh, 2000) is based on identifying varieties of rice using 8e10 microsatellites (simple sequence repeats) by sizing them on a capillary sequencer. However, lab-on-a-chip has less discriminating power than capillary sequencer.

The assay was redesigned using (insertion or deletion of base pair sequences) markers to give a method, which although less able to identify individual varieties of rice, is a more practical screening test for Basmati adulteration

PROTEOMIC METHODS

Peptide and protein sequence data are combined with the analytical capabilities of mass spectrometry (MS) in a

relatively recent proteomics method. Pep-tides from protein digests with as low as 108,8 mol of protein can now be detected and identified thanks to recent developments in MS technologies. Prior to doing the peptide separations online, protein fractionation can be carried out offline to increase the power of MS detection.

Myofibrillar peptides are particularly noteworthy because they remain largely unaffected by cooking or heat processing. The technique that was developed could identify 0.5% w/w of a contaminating meat species within another meat species, even after it has been cooked (Sentandreu, Fraser, Halket, Patel, & Bramley, 2010). An additional application has been to identify and differentiate gelatin present in vegetarian food items, ensuring they are free from any animal-derived ingredients. A proteomic method utilizing acid hydrolysis successfully identified specific collagen peptide markers, in addition to markers for bovine and pork gelatin (Halket, Patel, Fraser, & Bramley, 2010). Another "shotgun" proteomic method involving tryptic digestion has also been utilized to differentiate gelatin, and a recent study conducted by the FSA combined both proteomic methods to identify hydrolyzed collagen proteins used in water- retaining agents for chicken products.

The shotgun proteomic technique was capable of measuring the mass of all tryptic peptides within a sample and subsequently comparing these peptides to sequence databases to identify the source of the hydrolyzed collagen proteins, instead of searching for a specific peptide marker linked to a single species (Collins et al., 2010; FSA study on injection powders utilized as water- retaining agents in frozen chicken breast products, 2009; Helen Grundy and Paul Reece, Food and Environment Research Agency & Matthew Collins, University of York, unpublished observations). In this research, the use of proteomics methods facilitated the analysis of highly processed samples where DNA was likely to be compromised.

METABOLOMICS

In metabolomics, the goal is to identify and measure as many small molecular weight compounds as possible. To ensure authenticity, it is important that the genuine material is distinguishable from adulterants or substitutes based on the presence or lack of a specific low molecular weight analyte.

There has been a longstanding need to identify mechanically separated meat (MSM), also referred to as MRM, in meat products to enforce specific regulations concerning the production and labeling of MSM.

Over the last 15 years, a variety of methods have been attempted without success. All these approaches involved targeted analysis, focusing on detecting differences in specific compounds between MSM and hand-deboned meat.

Through gas chromatography-mass spectrometry (GC-MS), it was determined that the levels of certain metabolites significantly varied among MSM, hand-deboned meat, and desinewed meat.

These metabolites could serve as potential biomarkers. However, it proved unreliable to determine the source of new meat samples solely based on biomarker concentrations. This was especially true for meat mixtures, where the variability among different samples overshadowed the differences between meat types.

Nevertheless, success was achieved through a global approach in which all metabolites identified via GC-MS were analyzed using chemometrics. MSM was effectively distinguished from other meat sources and could be identified in raw meat mixtures at a concentration as low as 10% (Jiye et al., 2010).

This signifies a major technological advancement. Interestingly, it was discovered that histological analysis of stained samples is highly effective for assessing whether raw materials (in-factory) comply with the legal definition of MSM (Groves, 2010). Therefore, there exists the potential to integrate a cutting-edge technique (metabolomics) with a well-established method (histology) to deliver a conclusive analysis.

Techniques

Summary of adulterants and detection methods (Honey and Juices)

Types of adulterants such as urea, detergents, added water, and the blending of milks from different animals (for instance, cow, goat, sheep).

Detection tools comprise paper-based systems, smartphone colorimetry, PCR, and LIBS (Laser- Induced Breakdown Spectroscopy).

Detection methods vary from colorimetric techniques for chemical adulterants to qPCR for distinguishing milk from various species, and LIBS combined with chemometric analysis for determining origin.

The research was conducted in India, Brazil, and Turkey, supported by relevant academic citations.

1. Cheese

Adulterants: Cellulose, silicon dioxide, wheat flour/semolina, and sawdust
Technique: Digital image analysis utilizing a Canon camera and multivariate analysis
Country: Argentina

Source: Visconti et al. (2023)

2. Butter

Adulterant: Lard fat

Technique: Raman spectroscopy combined with chemometric methods
Country: Turkey

Source: Taylan et al. (2020)

3. **Milk Powder**

Adulterant: Nonprotein nitrogen

Technique: Raman spectroscopy and point-scan imaging
Country: China

Source: Yang et al. (2022)

4. **Milk Powder**

- Contaminants: Rice flour and soy flour
 - Technique: FTIR (Fourier Transform Infrared) spectroscopy employing chemometric approaches
- Country: China

• Source: Feng et al. (2019)

Summary of meat and meat products

1. **Beef mixed with offal**

- Technique: UV-Visible spectroscopy, fluorescence spectra, and multispectral imaging (MSI)
- Country: Greece

Source: Fengou et al. (2021)

2. **Pork mixed with chicken**

- No distinct method specified, presumably included in the multispectral analysis mentioned earlier.

3. **Minced beef mixed with lamb, pork, poultry, duck meat, and textured vegetable protein (TVP)**

- Method: Color imaging system (LED and camera) combined with deep neural network
- Country: Indonesia

Source: Setiadi et al. (2022)

5. **Minced beef mixed with pork**

- a. Method: RT-PCR using Ladder-shape melting temperature isothermal amplification (LMTIA)
- Country: China

6. **Minced Beef mixed with Pork**

- Technique: RT-PCR utilizing Ladder-shape melting temperature isothermal amplification (LMTIA)
- Location: China
- Citation: Yongzhen et al. (2022)

7. **Sheep meat mixed with Chicken and Duck**

Method: PCR and high-efficiency identification (details partially omitted)

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- Location: China
- Citation: Yang et al. (2022)

Detection Techniques (Fruits & Vegetables)

1. **Apples and Oranges Adulterant: Formaldehyde**

Method: HCHO gas sensor connected to Raspberry Pi3, utilizing machine learning
Country: India

Reference: Brighty et al. (2021)

2. **Carob, Fig, Grape, Mulberry Adulterants: Glucose and fructose syrup**

Method: Elemental Analyzer-Isotope Ratio Mass Spectrometry (EA-IRMS) with statistical analyses
Country: Turkey

Reference: Yücel et al. (2023)

3. **Bitter Gourd, Okra, Pointed Gourd Adulterant: Copper sulfate**

Method not specifically mentioned (likely traditional chemical marker detection or visual analysis)

4. **Red Chilies**

- Contaminant: Sudan red dye
- Technique: Low-field nuclear magnetic resonance (NMR) spectroscopy
Country: United States
- Source: Shomaji et al. (2021)

5. **Yellow and Green Peas**

- Contaminant: Malachite green
- Technique: NMR relaxometry coupled with chemometric analysis
Country: United States
- Source: Shomaji et al. (2021)

Summary of Adulterants and Detection Techniques (Grains & Flour)

1. **Black Rice mixed with Different Rice Types**

- Technique: Near-Infrared Spectroscopy (NIRS) utilizing data-driven class modeling and variable selection

Country: China

• Source: Chen et al. (2018)

2. **Rice mixed with other Rices**

- Technique: Use of a photographic camera together with Convolutional Neural Networks (CNNs) for analyzing images

• Country: Spain

- Reference: Izquierdo et al. (2020)
3. ***Corn contaminated with Multiclass Pesticide Residues***
 - Technique: Capillary electrophoresis tandem mass spectrometry combined with QuEChERS
 - Country: Brazil
 - Reference: Daniel and do Lago (2019)
 4. ***Durum wheat in flour and pasta mixed with common wheat***
 - Technique: Laser-Induced Breakdown Spectroscopy (LIBS) utilizing chemometrics
 - Country: Turkey
 - Reference: Sezer et al. (2022)

Summary of Adulterants and Detection Methods (Oils & Fats)

1. ***Organotin has been found to adulterate sesame, olive, peanut, soybean, and corn oils.***
 - Technique: Capillary electrophoresis-electrospray ionization-mass spectrometry (CE-ESI- MS)
 - Location: China
 - Citation: He et al. (2020)
2. ***Sunflower and canola oils have been used to adulterate cod liver oil.***
 - Equipment: NMR spectrometer, FTIR spectrometer, GCMS spectrometer
 - Methods: 1H and 13C NMR spectroscopy, FTIR, GCMS combined with Artificial Neural Networks (ANN)
 - Location: Germany
 - Citation: Giese et al. (2019)
3. ***Camellia oil mixed with corn, rapeseed, and sunflower oils***
 - Technique: Near-infrared (NIR) spectroscopy using chemometric analysis Country: China
 - Citation: Du et al. (2022)
4. ***Verification of peanut oil authenticity***
 - Technique: Raman spectroscopy combined with chemometric analysis Country: China
 - Citation: Peng et al. (2022)

Summary of Adulterants and Detection Methods (Honey & Juices)

1. ***Genuine honey mixed with commonly***
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adulterated honey

- Technique: Electronic tongue combined with chemometrics
 - Location: Colombia
 - Source: Leon-Medina et al. (2023)
2. ***Honey combined with fructose, glucose, inverted sugar, hydrolyzed inulin syrup, and malt must***
 - Technique: Raman spectroscopy with chemometrics
 - Location: Romania
 - Source: Oroian et al. (2018)
 3. ***Honey from Suichang that has been mixed with general honey***
 - Technique: Raman spectroscopy using machine learning algorithms Country: China
 - Reference: Hu et al. (2022)
 4. ***Unadulterated honey mixed with molasses, date molasses, grape molasses, high fructose corn syrup, corn syrup, sucrose, and inverted sugar***
 - Technique: Raman spectroscopy with pattern recognition analysis Country: USA
 - Reference: Aykas et al. (2020)

Applications

- Food forensics involves the scientific examination of food to guarantee its safety, authenticity, and traceability. Here are some concise applications of food forensics:
 - Food Fraud Detection: Identifies instances of mislabeling or ingredient substitution (e.g., horse meat in beef products, counterfeit honey or olive oil).
 - Contaminant Identification: Identifies harmful agents such as pesticides, toxins, heavy metals, or pathogens present in food.
 - Traceability and Origin Verification: Verifies the geographic or species origin of food (e.g., distinguishing between wild and farmed fish, or confirming the country of origin for wines).
 - Genetically Modified Organism (GMO) Detection: Confirms the presence or absence of genetically modified components in food products.
 - Allergen Verification: Ensures that items are free from undeclared allergens that could be harmful to sensitive individuals.

- **Quality Control and Assurance:** Assists in maintaining consistent quality by identifying spoilage, adulteration, or alterations resulting from processing.
- **Legal and Regulatory Compliance:** Aids law enforcement and food agencies in examining and prosecuting food-related offenses.

CONCLUSION

The continuously growing global food supply chain offers both unique opportunities and significant challenges in maintaining food safety, authenticity, and consumer trust. This study has explored the interdisciplinary area of food forensics, highlighting the crucial role that forensic science plays in protecting public health and economic integrity. By thoroughly investigating scientific techniques—ranging from DNA testing and isotope ratio analysis to metabolomics and proteomics—it is clear that contemporary forensic methods are essential for detecting food fraud, contamination, adulteration, and mislabelling.

The research has underscored how the integration of conventional analytical chemistry with innovative technologies like CRISPR diagnostics, portable colorimetric sensors, and machine learning is transforming the identification and monitoring of food quality. Food forensics not only tackles fraudulent practices for financial benefit but also reveals significant ethical issues related to deliberate poisoning, biological contamination, and antibiotic resistance, particularly from livestock. The historical development of food forensics, spanning from early legal frameworks to contemporary scientific methods, illustrates that the quest for accountability and fairness in food systems is both enduring and critical.

Furthermore, this study has highlighted the significant real-life consequences of food borne diseases and the vital necessity for precise food labelling and traceability. The combination of forensic techniques with regulatory systems by organizations like the FDA, EFSA, and WHO signifies the increasing acknowledgment of food forensics as a crucial field in the battle against global food-related crimes.

In summary, food forensics serves as a protector of food quality in the modern era. Its significance is anchored not just in science and technology but also in the ethical responsibility to safeguard consumers, build public trust, and enhance global food security. As advancements in analytical methods progress, this field is set to provide even higher levels of accuracy, dependability, and agility. The future of food forensics depends on its capacity to respond to new challenges while upholding fundamental principles of safety, honesty, and fairness in every meal we consume.

DISCUSSION

The future of food forensics is highly promising as the need for safer, more transparent, and ethically produced food

continues to grow. As globalization increases food trade and supply chains become more intricate, the demand for quick, dependable, and portable forensic tools is more urgent than ever. This field is set to become essential for public health, consumer protection, and sustainable farming practices. One particularly exciting area for growth is the incorporation of artificial intelligence (AI) and machine learning into food forensic methods. These innovations can improve data analysis, uncover patterns of deception, and even forecast potential contamination risks throughout the supply chain. The advancement of AI-driven smart sensors and blockchain-based traceability systems will enable real-time monitoring and verification of food origin, composition, and safety.

Furthermore, portable and on-site detection systems like handheld DNA analyzers, colorimetric sensors, and microfluidic lab-on-a-chip devices will transform food inspection by enabling forensic analysis to be conducted in the field, at markets, and in processing units. This capability will be particularly beneficial in areas where access to advanced laboratory facilities is limited. The use of next-generation sequencing (NGS) and metabolomic profiling will enhance the detection of subtle adulterations and emerging contaminants. These technologies can be employed to monitor antibiotic resistance genes, identify novel pathogens, and analyze intricate food matrices with high sensitivity and specificity.

Additionally, international cooperation and the standardization of food forensic regulations will be essential for effectively addressing global food fraud. As food-related crimes cross international borders, there is an increasing necessity for universal guidelines, shared databases, and coordinated law enforcement efforts. Developing specialized units focused on food crime and investing in forensic resources will play a key role in this initiative. Furthermore, educational programs and skill enhancement in food forensics are expected to expand, with universities and research institutions creating specialized courses that integrate food science, analytical chemistry, and forensic investigation. This multidisciplinary approach will prepare a new generation of professionals to confront the changing challenges related to food authenticity and safety.

In the end, food forensics will safeguard consumers from financial and health risks while also promoting ethical practices, environmental sustainability, and social responsibility within food systems. The future of this field is marked by ongoing innovation, collaboration, and influence, establishing it as a crucial area in our joint quest for food integrity.

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