Eurasian Journal of Analytical Chemistry
ISSN: 1306-3057 OPEN ACCESS 2021 16 (2): 10-14

Recent Applications of Hyphenated Liquid Chromatography Techniques in Food Forensics

Dr. Sachil Kumar

Received: 02 February 2021 ▪ Revised: 28 February 2021 ▪ Accepted: 15 March 2021

Abstract: Typically, hyphenated techniques coupled with mass spectrometry are considered as robust and reliable confirmatory approach. This technique can be used for selective analysis of predefined adulterant in the food item, whereby food can be examined either using LC-MS or GC-MS with further validation of component configuration using NMR. This review article addresses the application of various techniques of hyphenated liquid chromatography used in food forensics.

Studies related hyphenated, liquid chromatography, food forensics, applications, mass spectrometry, and forensic science in various combinations were retrieved from systematic web search of "MEDLINE", "ScienceDirect", "PubMed" and "Google Scholar" databases with the last search performed in January 2020.

Hyphenated techniques such as LC-API-MS/MS can point to the exact origin of the blood or the issue of mixing species. LC-MS/MS in combination with NMR can be used to monitor certain food items for adulteration.

Keywords: Hyphenated Chromatography, Food Forensics, Analysis, Liquid Chromatography, Mass Spectrometry.

INTRODUCTION

New methodological possibilities have been uncovered by the innovative introduction of hyphenated methods for the forensic scientist. In recent years, hyphenated approaches have gained wide acceptance as the principal way of addressing complicated analytical issues [1]. Hyphenated intensity facilitates the detection and quantification of complex mixtures. Even the unidentified specimen may be examined with accuracy and specificity [2-3]. For the purpose of obtaining extensive structural information for the elucidation of analytes contained in rudimentary samples, separation techniques such as LC, HPLC, GC or CE are fused with spectroscopic detection technologies like FTIR, MS, NMR and PDA, leading to the introduction of numerous hyphenated methods such as GC / MS, CE / MS, LC / NMR and LC / MS [4]. In 1980, Thomas Hirschfield coined the word "hyphenation" to describe a possible online combination of separation and spectroscopic analytical methods in a single run cycle [5]. The relation between spectroscopy and liquid chromatography is essentially a physical link. LC-MS was used more commonly than LC-NMR, with greater sensitivity. Hyphenation may not necessarily have to be within 2 technologies; the integration of separation and detection technologies may include multiple separation or detection technologies, like LC/NMR/MS, LC/PDA/NMR/MS, and so forth. The online combination of SPE, SPME or LVI can be integrated into a more powerful embedded device, e.g. (LVI)-GC-MS or (SPE)-LC-MS, where trace analysis is essential and analyte enrichment is necessary [6].

Hyphenated methods have a high potential in food forensic. Food analytical methods increasingly require the development of more reliable, accessible, flexible and analytical methodologies to ensure food authentication, quality control and standardization in accordance with consumer requirements and legislation. Food analytical laboratories are now being enforced to replace their existing technologies with modern analytical techniques that enable them to respond appropriately to international standards for food safety, quality and traceability [8].

This article emphasis on a discussion upon the extensive involvement of advances hyphenated liquid chromatographic techniques in different kind of food forensic analysis.

Dr. Sachil Kumar, Department of Forensic Sciences, College of Criminal Justice, Naif Arab University for Security Sciences, Saudi Arabia. E-mail: skumar@nauss.edu.sa

MAIN TEXT

Hyphenated Techniques in Liquid Chromatography [7]

1. Double hyphenated analytical methods (LC/MS; LC/IR; LC/NMR)
2. Triple hyphenated analytical methods (LC/MS/MS**;** LC-API-MS; LC/APCI/MS/MS; LC/ESI/MS/MS: LC/UV/NMR/MS/ESI; LC/DAD/API/MS; LC/DAD/ESI/MS/MS; LC/PDA/NMR/MS; LC/SPE/NMR/MS)

Identification of Related Studies

We performed a systematic web search of “MEDLINE”, “Sciencedirect”, “PubMed” and “Google Scholar” databases for all the related publications using the keywords hyphenated, liquid chromatography, food forensics, applications, mass spectrometry, and forensic science in various combinations. Broad search terms were used to assist the identification of all pertinent articles, with the last search performed in January 2020. All the relevant studies have been described in following section.

Food Forensics

The term "food forensics" implies the use of specialized science to investigate food crimes. It has been used to address food theft and adulteration problems, allegations of mislabeling or fraud, impose supervision or control, and resolve other legal problems related to food goods and ingredients' health and authenticity. Food adulteration in the past few years may have major consequences for people's health and can hinder business growth by losing consumer trust. Food Adulteration refers to the process by which the consistency or value of a given food is decreased by expanding adulterants or evacuating critical substances. Food forensics is more generally a collection of methods that provide revolutionary traceability across the supply chain and these techniques are used not only for food but also in pet and pharmaceutical products [9].

Food Forensic analysis is used to determine what, how and where food safety and consistency problems concerning the detection of food authenticity or certification of foreign matter, taints and odors. Food Forensics inquiries are carried out for a variety of reasons, including environmental safety, corporate accountability and reputation, regulatory criteria. The risk of food being deliberately or accidentally altered or contaminated also increases as already growing variety of foods originate from more diverse sources and are processed in a wide variety of ways. Meat forensicists are thus facing significant and advanced challenges. One that method of exploitation is the commercial replacement of more expensive ingredients with low priced but identical food. Seafood harvested all across the globe and frequently sold in remote areas far from its harvest is often inadvertently or knowingly mislabeled [10]. The identification of both farmed and wild-harvested species such as salmon is more complicated. Less costly horse meat was mislabeled as comparatively costlier beef or pork in all of Europe [11]. Through infant milk laced with melamine [12] and horse meat patties as adulteration of beef [13] products is popular, but there is still a gap between production, consumption and regulatory oversight to keep food fraudsters in check.

There are most notable incidents linked to food adulteration, such as the 2013 meat adulteration scandal, also known as the Horse Gate, where unrevealed horse meat was disguised as branded beef in food products, where about 30 000 bottles of adulterated low quality wine labeled as Brunello di Montalcino, Chianti, are other popular Italian wine scams. Eventually, the so-called Honey Laundering Scheme dubbed 'the biggest food scam in U.S. history’, where honey manufactured in China was sold on the U.S. market via a web of Asian nations to 'cover' fraudulent labels and fabricated evidence for the Chinese origin commodity. Without a question Italy is one of the countries most affected by food safety issues. Melamine was used in China to mislead the Kjeldahl-based protein analysis so that milk appears more condensed than it would have been.

In 2008, the Chinese authorities found that numerous Chinese manufacturers used melamine to adulterate milk and infant formula. Hundreds of thousands of injuries and six deaths have been reported in China, as have product recalls in other countries [14]. Turmeric manufactured in India or Bangladesh and sold in the USA has been found to be adulterated with lead (II) chromate in order to increase color and weight [15]. Ciguatoxin 1B is one example of a typical, structurally complex aquatic poison that can naturally pollute a variety of seafoods [16]. Aside from harmful chemicals, food may be polluted with other toxins from small molecules, intoxicating proteins (prions), protein contaminants or toxin-producing bacteria [17]. Additives such as transglutaminase, commonly referred to as meat glue, are licensed for use in binding meat trimmings and off-cuts together; however specific food attachments have

also been identified in cases of fraud where perpetrators have attempted to implement them with a view to intentionally raising the meat content on labels [18-19].

Utilization of LC-MS Hyphenated Techniques in Food Forensic

In addition to basic understanding, the sustainability, reliability and efficiency of the existing food network worldwide depends heavily on food analysis as a tool for the implementation of new goods, product testing, trade regulations and significant problem solutions [20]. Targeted analyzes based on identification and detection is the most widely used analytical methods. Recognition of a specific molecule or category of compounds (amino acids, organic acids, sugars, etc.); such components may or may suggest malpractices as authentic biomarkers of the product [21]. The holistic technique ensures accurate and sensitive analysis and appropriate quantification of different compounds, and can be used to test specific features of substances or variables that reflect the sample or its spoilage and its route of origin [22].

There are several possibilities of hyphenated approaches being implemented in food forensics [23]. Coupling of liquid chromatography with mass spectrometry (LC-MS) or tandem MS (LC-MS/MS) commonly used for food safety [24-25], especially for the study of antimicrobial residues in foodstuffs of animal origin [25-26], antibiotics in food samples [27], clenbuterol residues [28], food allergens [29] and so forth. The gradually growing need of LC hyphenated with MS (HRMS) analyzers offers a large involvement of LC MS-based authenticity approaches that delivers best outcome from authenticated food analysis [30]. Citrus polyphenolic profiles have also been analyzed in detail to distinguish Green, Mandarin, Grapefruit and Lemon juices. Abad Garcia et al. proposed a comprehensive analysis to evaluate 49 polyphenols in citrus juices using HPLC-DAD-ESI-MS/MS [31].

Grundy and others (2007, 2008) LC-API-MS (/MS) developed and validated to differentiate between the fibrin clots used in binding procedures. Fibrio-peptides are species-specific in respect of their molecular weights, especially considering that fragments up to or greater than MS3 formed in LC-API-MS(/MS) ensure that even similar peptides in a given MS-MS experiment have differences in the fragmentation. in a given MS-MS experiment. LC-API-MS(/MS) can provide information about the origin or source of blood in combination of different species [18-18]. LC/MS/MS is used in combination with NMR to validate cases of spoilage in other food items, such as monofloral honey due to the presence of rare and highly sensitive biomarkers in the nectar of plant species used by bees. [32]. LC/MS/MS framework was proposed to confirm the possible existence of peanut in chilli peppers [33]. LC-MS will complement current methods for detecting adulterated juices in conjunction with chemo-metric examination [34].

Polyphenols are a major category of naturally derived secondary metabolites of plant-derived foods which related to micro nutrients or beneficial health effects needed by a healthy diet. The different chemical configuration of polyphenols has impeded the analyte from being extracted and processed, as well as its isolation, recognition and determination.

The LC-MS or LC-MS/MS is the most popular approach for structural configuration and determination of low to high molecular weight polyphenols in food samples [35]. In recent years, High-resolution mass spectrometry (LC-HRMS) methods have played an essential part in polyphenol research, either by using time-of-flight (TOF) or by characterizing orbitrap analyzers, not just in evaluating this compound family in foodstuffs, but also in classifying and identifying unknown polyphenols.

CONCLUSIONS

Fraudulent food items, such as practicing falsification for cheaper products and misleading claims of origin (natural source), decrease the value of the products; the consumers deceives and may even pose a significant health threats. Consequently, for all those engaged in food trade, food quality is a potential concern: purchasers, public health authorities, as well as suppliers and dealers. More rapid and precise analytical tools are now used by field experts to measure and monitor food adulterants that are both simplified and price-effective. LC–MS–MS and LC-API-MS(/MS) have developed into a powerful and accurate methods that provides flexibility, precision and responsiveness.

ABBREVIATIONS

LC: Liquid Chromatography; DAD: Diode Array Detection; API: Atmospheric Pressure Ionization; MS: Mass Spectrometry; ESI: Electro Spray Ionization Tandem; API: Atmospheric Pressure Ionization; PDA: Photodiode Array; NMR: Nuclear Magnetic Resonance; SPE: Solid-phase Extraction; TSP: Thermo Spray; UV: Ultraviolet; IR: Infrared Spectroscopy; HPLC: High-performance Liquid

Chromatography; CE: Capillary Electrophoresis; FTIR: Fourier-transform Infrared; SPE: Solid-phase Extraction; LVI: Large Volume Injection; SPME: Solid-phase Micro Extraction.

Competing Interests: None.

Funding: Nill.

REFERENCES

[1] Michalski R, Szopa S, Jabłońska M, Łyko A. Application of hyphenated techniques in speciation analysis of arsenic, antimony, and thallium. *The Scientific World Journal.* 2012 Jan 1; 2012.

[2] Mohamad Taleuzzaman and Sadaf Jamal Gilani. Brief on -Hyphenated Methods of HPLC for Determining the Presence of Solutes. *Chronicles of Pharmaceutical Science* 1.1 (2017): 35-42.

[3] Piotr M., *et al*. Application of high-performance liquid chromatography on-line with ultraviolet/visible spectroscopy in food science. *Polish journal of food and nutrition sciences* 15.56 (Suppl. 1) (2006): 145–153.

[4] Duncan WP. *Analytical Methods, Hyphenated Instruments.* Kirk-Othmer Encyclopedia of Chemical Technology. 2000 Dec 4.

[5] Wilson ID, Brinkman UT. Hyphenation and hypernation: the practice and prospects of multiple hyphenation. *Journal of Chromatography* A. 2003 Jun 6; 1000(1-2): 325-56.

[6] Andrews GR. Distinguishing pasteurized, UHT and sterilized milks by their lactulose content. *Journal of the Society of Dairy Technology* 37.3 (1984): 92–95.

[7] Nagajyothi S, Swetha Y, Neeharika J, Suresh PV, Ramarao N. Hyphenated Techniques-A Comprehensive Review. *International Journal for Advance Research and Development.* 2017;2(4).

[8] Alejandro Cifuentes, 2012, Food Analysis: Present, Future, and Foodomics, *International Scholarly Research Network ISRN Analytical Chemistry* Volume 2012, Article ID 801607, 16 pages https://doi.org/10.5402/2012/801607

[9] Maurizio Aceto, Food Forensics, *Comprehensive Analytical Chemistry,* Vol. 68. https://doi.org/10.1016/B978-0-444-63340-8.00009-1,2015.

[10] Stahl, A.; Schroder, U. Development of a MALDI-TOF MS-based protein fingerprint database of common food fish allowing fast and reliable identification of fraud and substitution. *J. Agric. Food Chem.* 2017, 65, 7519-7527.

[11] Walkera, M. J.; Burns, M.; Burns, D. T. Horse meat in beef products-species substitution 2013. *J. Assn. of Pub. Anal.*, (Online) 2013, 41, 67-106.

[12] http://www.who.int/csr/media/faq/QAmelamine/en

[13] https://www.bbc.co.uk/news/uk-21335872

[14] Fanny Fu, AB SCIEX Taipei (Taiwan), André Schreiber, AB SCIEX Concord, Ontario (Canada), Quantitation and Identification of Dicyandiamide in Milk and other Protein-Rich Foods, LC-MS/MS Analysis of Emerging Food Contaminants, 2013.

[15] Cowell, W.; Ireland, T.; Vorhees, D.; Heiger-Bernays, W. Ground turmeric as a source of lead exposure in the United States. *Public Health Rep.*, 2017, 132, 289-293.

[16] Caillaud, A.; De la Iglesia, P.; Darius, H. T.; Pauillac, S.; Aligizaki, K.; Fraga, S.; Chinain, M.; Diogene, J. Update on methodologies available for ciguatoxin determination: perspectives to confront the onset of ciguatera fish poisoning in Europe. *Mar. Drugs* 2010, 8, 1838-1907.

[17] Duracova, M.; Klimentova, J.; Fucikova, A.; Dresler, J. Proteomic methods of detection and quantification of protein toxins. *Toxins (Basel)* 2018, 10.

[18] Grundy HH, Reece P, Sykes MD, Clough JA, Audsley N, Stones R. Screening method for the addition of bovine blood-based binding agents to food using liquid chromatography triple quadrupole mass spectrometry. *Rapid Communications in Mass Spectrometry: An International Journal Devoted to the Rapid Dissemination of Up-to-the-Minute Research in Mass Spectrometry.* 2007 Sep 30;21(18):2919-25.

[19] Grundy HH, Reece P, Sykes MD, Clough JA, Audsley N, Stones R. Method to screen for the addition of porcine blood-based binding products to foods using liquid chromatography/triple quadrupole mass spectrometry. *Rapid Communications in Mass Spectrometry: An International Journal Devoted to the Rapid Dissemination of Up-to-the-Minute Research in Mass Spectrometry.* 2008 Jun 30; 22(12): 2006-8.

[20] R. J. Mcgorrin, One hundred years of progress in food analysis, *Journal of Agricultural and Food Chemistry,* vol. 57, no. 18, pp. 8076–8088, 2009

[21] Navarro-Pascual-Ahuir, M.; Lerma-García, M.J.; Simó-Alfonso, E.F.; Herrero-Martínez, J.M. Analysis of Aliphatic Organic Acids in Commercial Fruit Juices by Capillary Electrophoresis with Indirect UV Detection: Application to Differentiation of Fruit Juices. *Food Anal. Method* 2017, 10, 3991–4002.

[22] Borras, E.; Ferre, J.; Boque, R.; Mestres, M.; Acena, L.; Busto, O. Data fusion methodologies for food and beverage authentication and quality assessment—A review. *Anal. Chim. Acta* 2015, 891, 1–14.

[23] Alejandro Cifuentes, 2012, Food Analysis: Present, Future, and Foodomics, *International Scholarly Research Network ISRN Analytical Chemistry* Volume 2012, Article ID 801607, 16 pages. https://doi.org/10.5402/2012/801607

[24] A. K. Malik, C. Blasco, and Y. Pico, Liquid chromatography- ´ mass spectrometry in food safety, *Journal of Chromatography A,* vol. 1217, no. 25, pp. 4018–4040, 2010.

[25] N. Mochizuki, Problems on LC-MS/MS analysis to ensure food safety, *Journal of the Pharmaceutical Society of Japan,* vol. 131, no. 7, pp. 1019–1025, 2011.

[26] S. Bogialli and A. Di Corcia, Recent applications of liquid chromatography-mass spectrometry to residue analysis of antimicrobials in food of animal origin, *Analytical and Bioanalytical Chemistry,* vol. 395, no. 4, pp. 947–966, 2009.

[27] M. C. Moreno-Bondi, M. D. Marazuela, S. Herranz, and E. Rodriguez, An overview of sample preparation procedures for LC-MS multiclass antibiotic determination in environmental and food samples, *Analytical and Bioanalytical Chemistry,* vol. 395, no. 4, pp. 921–946, 2009.

[28] J. Pleadin, I. Bratos, A. Vuli ˇ c, N. Per ´ si, and J. Dugum, Analy- ˇ sis of clenbuterol residues in pig liver using liquid chromatography electrospray ionization tandem mass spectrometry, *Reviews in Analytical Chemistry,* vol. 30, no. 1, pp. 5–9, 2011.

[29] C. K. Faeste, H. T. Rønning, U. Christians, and P. E. Granum, Liquid chromatography and mass spectrometry in food allergen detection, *Journal of Food Protection,* vol. 74, no. 2, pp. 316–345, 2011.

[30] Ibáñez, C.; García-Cañas, V.; Valdés, A.; Simó, C. Novel MS-based approaches and applications in food metabolomics. *Trac-Trend Anal. Chem.* 2013, 52, 100–111.

[31] Marilena E. Dasenaki * and Nikolaos S. Thomaidis, Quality and Authenticity Control of Fruit Juices-A Review, Molecules 2019, 24, 1014; https://doi.org/10.3390/molecules24061014.

[32] Wang Z, Jablonski JE. Targeted and non-targeted detection of lemon juice adulteration by LC-MS and chemometrics. *Food Additives & Contaminants: Part A.* 2016 Mar 3; 33(3): 560-73.

[33] Donarski JA, Roberts DP, Charlton AJ. Quantitative NMR spectroscopy for the rapid measurement of methylglyoxal in manuka honey. *Analytical Methods.* 2010; 2(10).

[34] Vandekerckhove M, Van Droogenbroeck B, De Loose M, Taverniers I, Daeseleire E, Gevaert P, Lapeere H, Van Poucke C. Development of an LC-MS/MS method for the detection of traces of peanut allergens in chili pepper. *Analytical and Bioanalytical Chemistry.* 2017 Sep 1; 409(22): 5201-7.

[35] H.J. Li, M.L. Deinzer, Tandem mass spectrometry for sequencing proanthocyanidins. *Anal. Chem.* 79 (2007) 1739-1748.