

Exploring the potential of metabolomics in the dairy industry: A mini review

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Abstract

The dairy industry is crucial for meeting global food and nutrition demands, providing high-quality proteins, calcium, vitamins, and minerals. However, the image of milk as white ambrosia has been tarnished due to adulteration, which poses risks to consumer health and compromises the integrity of dairy products. Traceability in the milk and dairy product industry ensures the safety, quality, and transparency of the entire supply chain. This allows for meticulous tracking of each stage of production and distribution, enabling quick identification of potential issues and addressing them. Metabolomics is a rapidly advancing field of study that focuses on the comprehensive analysis of small molecules or metabolites within a biological system. It involves identifying, quantifying, and analyzing these metabolites to understand the metabolic processes occurring in cells, tissues, and organisms. Metabolomics has been used in numerous applications in the dairy industry, providing valuable insights and aiding in various aspects of production, quality control, and product development. By harnessing the power of metabolomics, the dairy industry can improve quality control, enhance animal health and welfare, develop innovative products, and meet the ever-evolving demands of consumers.

Keywords: Authentication, Mass spectroscopy, NMR, Quality

Highlights

- Metabolomics, a crucial aspect of omics sciences, has experienced significant growth in recent years, especially in its application within the dairy industry.
- This mini-review provides a comprehensive analysis of metabolomics's role in the dairy industry.
- Metabolomics offers valuable insights into the processes influencing milk production, composition, and quality.
- The paper explores the use of metabolomics in assessing milk bioactivity, thereby improving the nutritional value and safety of dairy products.
- Metabolomics holds great potential to improve dairy products' quality, safety, and nutritional content.

INTRODUCTION

The dairy industry plays a vital role in meeting global demands of major foods and essential nutrition. Dairy products derived from milk provide a rich source of high-quality proteins, calcium, vitamins and minerals, making them an important component of a balanced diet (Górska-Warsewicz *et al.*, 2019). Moreover, dairy products contribute to food security by providing a reliable and accessible source of nutrition, particularly in regions where alternative sources of protein and essential nutrients are scarce (Chaudhary and Krishna, 2021). Therefore, it is considered as almost complete food or natural ambrosia.

However, the image of milk as white ambrosia to human beings, has been tarnished due to the curse of adulteration in milk. Milk adulteration, involving unethical practices such as dilution with water, use of

low-quality ingredients, or the addition of synthetic additives, poses risks to consumer health and compromises the integrity of dairy products (Azad and Ahmed, 2016; Handford *et al.*, 2016). However, apart from adulteration, contamination (chemical and microbiology) is a major threat to the quality of dairy products. Foodborne pathogens, microbial spoilage, and the presence of mycotoxins can pose significant health risks for the consumer. Consumers today are more conscious about the origin and quality of their food and traceability provides them with the assurance that the milk and dairy products they consume adhere to stringent standards, fostering trust in the industry and ensuring a safer and more accountable dairy supply chain.

Traceability, in the milk and dairy product industry is a crucial aspect that ensures the safety, quality, and transparency of the entire supply chain. From the farm

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to the consumer's table, traceability allows for the meticulous tracking of each stage of production and distribution. Comprehensive traceability not only enables quick identification of any potential issues or contamination but also helps swiftly address and resolve them. Therefore, the dairy industry is always in search of innovative technologies that provide comprehensive information regarding product safety, quality and traceability.

In the dairy industry, metabolomics offers a powerful tool to investigate the composition, quality and safety of milk, as well as to understand the metabolic processes occurring during milk production and processing. Overall, the use of metabolomics in the dairy industry has revolutionized our understanding of milk composition, animal metabolism, product quality, and product development (Rocchetti and O'Callaghan, 2021; Suh, 2022). By harnessing the power of metabolomic analyses, the dairy industry can improve quality control, enhance animal health and welfare, develop innovative products, and meet the ever-evolving demands of consumers. Therefore, this mini-review paper aims to provide a comprehensive overview of the scope and applications of metabolomics in the field of milk or other dairy product production or processing.

Fundamentals of metabolomics

The term "omics" is derived from the Greek word "ome," which means "all" or "every." It is used as a suffix in various scientific disciplines to denote a comprehensive or global approach to studying a particular aspect of biology. Omics fields involve analyzing large-scale biological data to gain a holistic understanding of biological systems. The most notable examples of "omics" are genomics, transcriptomics, proteomics, and metabolomics (Debnath *et al.*, 2010).

Metabolomics is a rapidly advancing field of study that focuses on analyzing small molecules (<1500 Da) or metabolites within a biological system (Muthubharathi *et al.*, 2021). It aids in understanding metabolic processes in cells, tissues, and organisms. Metabolomics has been used in dairy industry applications, providing insights into production, quality control, and product development (Suh, 2022; Wu *et al.*, 2022). It offers a holistic approach to understanding animal interactions. (Sen *et al.*, 2021).

Targeted metabolomics and untargeted metabolomics are complementary approaches in metabolomics research, focusing on quantitative analysis of predefined metabolites and comprehensive profiling without a specific target. Targeted metabolomics provides precise quantification, allowing in-depth investigations of metabolic pathways (Roberts *et al.*, 2012).

Untargeted metabolomics provides a holistic view of the metabolome, enabling researchers to discover novel biomarkers and metabolic signatures, enabling targeted investigations and exploratory studies. (Schrimpe-Rutledge *et al.*, 2016). Table 1 shows the difference between Targeted and untargeted metabolomics.

Basic principle of metabolomics

A typical metabolomic study involves several key steps, including sample collection, analytical platforms like mass spectrometry (MS) or NMR spectroscopy, data pre-processing, and analysis and interpretation (Chen *et al.*, 2022) (Fig. 1). Sample handling and processing are crucial to preserve metabolite composition and prevent degradation. MS-based metabolomics offers high sensitivity and enables the identification and quantification of a wide range of metabolites, while NMR provides

Table 1. Comparison of targeted and untargeted metabolomics

Aspects	Targeted metabolomics	Untargeted metabolomics
Objective	Quantify specified pre-selected metabolites	Profile a wide range of metabolites without pre-selection
Application	Well-suited for validation and targeted studies	Suitable for biomarker discovery, system-level study
Scope	Narrow, focused analysis	Broad, comprehensive analysis
Sensitivity	High sensitivity	Low sensitivity, may miss low-abundance or unexpected metabolites
Quantification	More accurate quantification of targeted metabolomics, requires purified standard	Less accurate due to an unknown compound
Sample	Small sample sizes sufficient	Require larger sample sizes due to variability in the metabolome

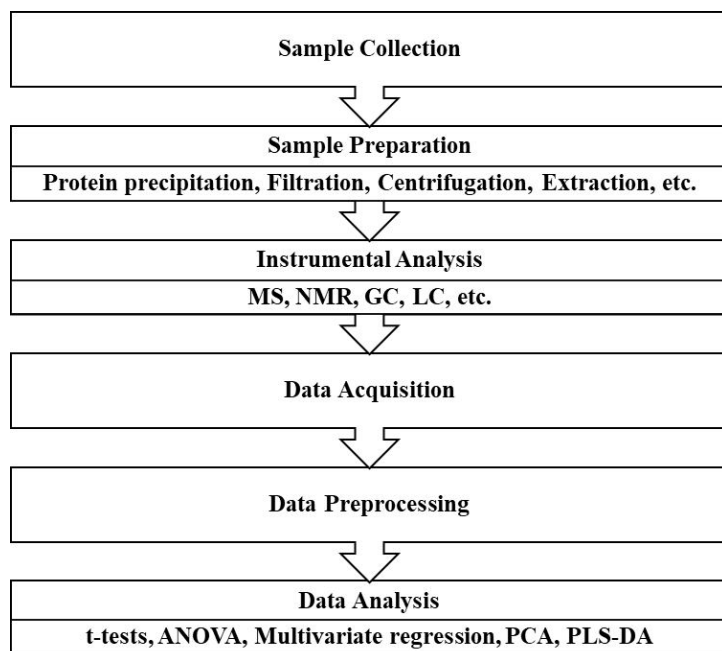


Fig. 1. Basic process of metabolomic analysis

non-destructive and quantitative analysis (Sen *et al.*, 2021). These techniques are often combined with separation techniques like liquid chromatography or gas chromatography. Data pre-processing steps remove noise, correct for systematic variations, and align data across multiple samples.

Metabolomic studies utilize statistical techniques, multivariate analysis, and bioinformatics tools to extract meaningful information from complex datasets. Advanced approaches like spectral databases aid in interpretation and studies are validated through experimental validation and integration with other omics data (Chen *et al.*, 2022).

Analytical methods used in metabolomics

Nuclear magnetic resonance (NMR) spectroscopy:

Nuclear magnetic resonance (NMR) spectroscopy is a widely used technique in metabolomic studies of milk and milk products. NMR offers several advantages, including its non-destructive nature, high reproducibility, and ability to provide both qualitative and quantitative information about metabolites (Emwas *et al.*, 2019). NMR-based metabolomics enables the simultaneous detection and quantification of a wide range of metabolites present in milk, including sugars, organic acids, amino acids, lipids, and volatile compounds (Sundekilde *et al.*, 2013). This comprehensive profiling provides valuable insights into the nutritional quality, authenticity, and processing effects of milk. The non-destructive nature

of NMR allows for repeated measurements on the same sample, facilitating longitudinal studies and reducing sample variability (Nagana Gowda and Raftery, 2021).

Proton nuclear magnetic resonance (^1H NMR) spectroscopy is a widely used technique in milk metabolomic research, allowing for the identification and quantification of various metabolites in milk. It has been used to investigate compositional differences in milk from different animal species, factors like diet, breed, and lactation stage, and to assess the effects of processing methods like pasteurization and fermentation on milk product metabolic profiles (Lamanna *et al.*, 2011; Sundekilde *et al.*, 2013).

Carbon-13 NMR spectroscopy provides information about the carbon atom environment in metabolites, allowing for structural details and metabolic pathways determination. It has been used to analyze the carbon distribution in milk components, such

as lactose, fatty acids, and amino acids, and to evaluate the authenticity and traceability of milk and dairy products by detecting isotopic ratios and carbon signatures (Sundekilde *et al.*, 2013).

Two-dimensional NMR spectroscopy offers enhanced resolution and structural information compared to ^1D NMR. Methods like correlation spectroscopy, hetero nuclear single quantum coherence, and total correlation spectroscopy (TOCSY) have been used to elucidate the complex metabolite composition of milk and milk products, enabling the identification of metabolite connectivity, chemical shifts, and molecular structures, facilitating the characterization of minor metabolites and metabolite mixtures in milk (Vasavi *et al.*, 2011; Sundekilde *et al.*, 2013).

NMR spectroscopy provides structural information about metabolites, aiding in their identification and characterization. Researchers can accurately identify and quantify specific metabolites by comparing spectra of milk samples with databases or using spectral patterns (Smolinska *et al.*, 2012). This information can study variations in metabolite composition among different milk types or assess the effects of factors like feeding regimes or processing techniques. NMR-based metabolomics offers good reproducibility and high sensitivity, making it a reliable platform for analyzing complex matrices and detecting subtle changes in metabolite profiles. However, it has limitations, such as low throughput and challenging quantification in

complex mixtures like milk due to overlapping signals in NMR spectra (Pan and Raftery, 2007; Emwas, 2015).

Mass spectrometry: Mass spectrometry (MS) is a versatile analytical technique that plays a crucial role in metabolomic studies. Several MS-based techniques are employed to analyze the metabolites present in biological samples, including milk and milk products (Gowda and Djukovic, 2014). Liquid chromatography-mass spectrometry (LC-MS), Gas chromatography-mass spectrometry (GC-MS), Direct infusion mass spectrometry (DIMS), Matrix-assisted laser desorption/ionization (MALDI), High-resolution mass spectrometry (HRMS), etc. use for the studies of metabolomes (Gowda and Djukovic, 2014; Agin *et al.*, 2016).

Liquid chromatography-mass spectrometry (LC-MS) is a widely used MS-based metabolomics technique for milk analysis, enabling the separation, detection, and identification of various metabolites in complex samples (Lu *et al.*, 2008). It offers high sensitivity, comprehensive metabolome profiling, accurate mass measurement, tandem mass spectrometry, and comparison with spectral libraries. LC-MS is widely used for milk composition profiling, adulteration detection, processing effects evaluation, and characterization of milk from different species (Imperiale *et al.*, 2023).

GC-MS is another powerful MS-based metabolomics technique, offering excellent resolution and sensitivity for volatile and semi-volatile metabolites, particularly fatty acids, volatile compounds, and small polar metabolites. This technique has been used to analyse metabolites such as volatile fatty acids, organic acids, and lipid oxidation products in milk and dairy products (Putri *et al.*, 2022). GC-MS-based metabolomics has been utilized to assess the impact of various factors, such as animal feeding strategies, on the metabolite composition of milk, contributing to the understanding of milk quality and flavour characteristics (Rocchetti and O'Callaghan, 2021).

High-resolution mass spectrometry (HRMS) is a versatile technique that has gained popularity in milk metabolomic studies due to its ability to provide comprehensive metabolite profiling. HRMS, such as Fourier transform ion cyclotron resonance mass spectrometry (FT-ICR MS) or Orbitrap MS, enables accurate mass measurement and identification of a wide range of milk metabolites. HRMS has been utilized to explore the metabolic changes occurring during milk processing, microbial fermentation, and storage,

providing insights into the transformation of metabolites and the formation of bioactive compounds (Senyuva *et al.*, 2015). Additionally, HRMS-based metabolomics has been used to identify potential biomarkers related to milk quality, safety, and authenticity (Selamat *et al.*, 2021).

However, MS-based metabolomics techniques have limitations, such as the time-consuming and labour-intensive nature of extensive sample preparation procedures and the complexity of MS data analysis and interpretation. Additionally, ion suppression or enhancement effects may affect the accurate quantification of metabolites, making method validation and appropriate internal standards crucial for reliable and reproducible results (Dettmer *et al.*, 2007; Sen *et al.*, 2021).

Statistical analysis methods

Statistical analysis plays a crucial role in processing and interpreting metabolomic data obtained from various analytical techniques. It helps identify significant differences, discover patterns, and extract meaningful information from complex metabolite datasets. Various statistical analysis methods were used for data processing and interpreting, such as PCA, ANOVA, PLS-DA, Clustering, several metabolomic databases, etc. (Bartel *et al.*, 2013; Chen *et al.*, 2022) (Fig. 2).

Principal component analysis (PCA) is a widely employed multivariate statistical technique in metabolomics. PCA reduces the dimensionality of metabolite data and visualizes the overall patterns of similarity or dissimilarity among samples. It allows for the identification of inherent clustering, outliers, and trends in the data. PCA has been extensively used in metabolomic studies of milk and milk products to reveal compositional variations, identify potential biomarkers, and assess the effects of different factors on milk metabolite profiles (Liland, 2011; Bartel *et al.*, 2013; Ren *et al.*, 2015; Fakayode *et al.*, 2020).

Partial least squares-discriminant analysis (PLS-DA) is another powerful method utilized in metabolomics to classify and discriminate sample groups based on metabolite profiles. PLS-DA combines dimensionality reduction and class prediction to identify metabolites contributing to sample differentiation. It has been applied in milk metabolomic studies to differentiate milk samples from different origins, evaluate the impact of processing methods on metabolite composition, and identify biomarkers associated with milk quality and product authenticity (Liland, 2011; Ren *et al.*, 2015).

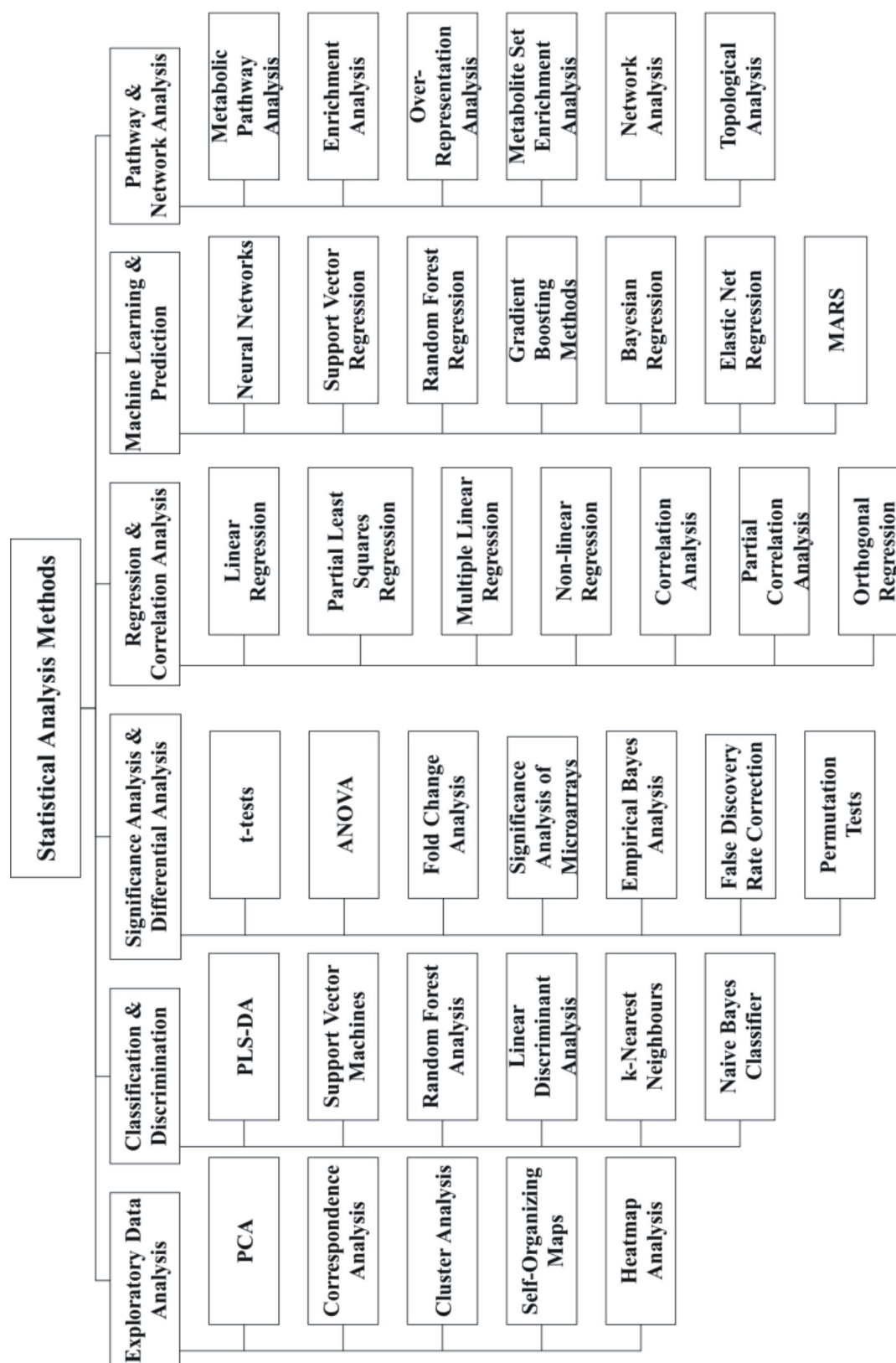


Fig. 2. Illustration of statistical analysis methods used in metabolomics

Hierarchical cluster analysis (HCA) is a commonly used unsupervised statistical technique that groups samples based on their similarity in metabolite profiles. HCA helps in identifying natural clusters and similarities within the data. It has been utilized in milk metabolomic studies to explore sample clustering based on factors such as breed, diet, lactation stage, and processing methods, providing insights into the underlying metabolic differences (Kellogg *et al.*, 2020).

Significant difference analysis, such as t-tests or analysis of variance (ANOVA), is employed to determine the statistical significance of differences in metabolite levels between different groups. These tests help identify metabolites that show significant changes related to factors of interest, such as treatment, genotype, or time points. Significant Difference Analysis has been used in milk metabolomic studies to identify metabolites affected by factors like breed, diet, or processing methods, contributing to the understanding of milk composition and quality (Yuan *et al.*, 2022; Zou *et al.*, 2022).

These statistical analysis methods are just a few examples of the diverse range of techniques employed in metabolomics to process and interpret complex metabolite datasets. The combination of multiple statistical approaches allows researchers to extract meaningful information, discover patterns, and identify potential biomarkers related to various factors in metabolomic studies (Bartel *et al.*, 2013). In addition to these methods, pathway analysis and enrichment analysis are employed to gain insights into the biological significance of the identified metabolites. These analyses utilize knowledgebases, such as the Kyoto Encyclopedia of Genes and Genomes (KEGG) (<https://www.genome.jp/kegg/>) or the MetaboAnalyst platform (<https://www.metaboanalyst.ca/>), MetAtt (<http://metatt.metabolomics.ca/>), Lipotype (<https://www.lipotype.com/>), etc. to map metabolites onto biochemical pathways, identify overrepresented pathways, and unravel metabolic networks associated with specific conditions or treatments.

Application of metabolomics in the dairy industry

Metabolomics is essential in the dairy industry for milk quality control, understanding the physiological and nutritional status of dairy animals, and developing functional dairy products. By analysing metabolite profiles, researchers can identify potential health-promoting components, enhance nutritional value, and improve animal welfare practices. Metabolomics also helps identify key flavour

components, enabling product optimization, flavour profiling, and maintaining consistency in product quality. By analysing volatile compounds and aroma profiles, metabolomics helps ensure the safety and authenticity of dairy products.

Metabolomics to analyse composition and nutritional quality: Metabolomics gives a new approach to determining the compositional parameters of milk and milk products. Several metabolites recognized in cow milk by ^1H NMR-based metabolomics concentrate on which lactose assumes an overwhelming part. ^1H NMR spectroscopy-based metabolomic studies were utilized for labelling and validation of milk nutrients and composition (Monakhova *et al.*, 2012). ^1H NMR is used to evaluate the lactase enzyme level, which is an important component of lactose digestion. These methods are utilized to evaluate the content of lactose in milk and milk items (Monakhova *et al.*, 2012).

In recent advancements within the field of milk analysis, Nuclear magnetic resonance (NMR) and Gas chromatography (GC) techniques have emerged as powerful tools for discerning the origins of different milk types based on their animal species. Andreotti *et al.* (2002) utilized ^{13}C -NMR spectroscopy, coupled with fuzzy logic analysis, to successfully differentiate bovine, sheep, and goat milk by examining their distinct fatty acid compositions. Additionally, Bruschetta *et al.* (2021) recently reported the efficacy of ^{31}P -NMR in discriminating milk samples from diverse animal species, irrespective of factors such as breed, age, or lactation. While Garcia *et al.* (2012) validate the approach of ^{13}C -NMR spectroscopy using Gas chromatography (GC) to distinguish milk samples from various animal species, showcasing its effectiveness through the identification and analysis of 12 different phospholipids (PL) profiles. In a comparative analysis of milk composition across diverse mammalian species, Wei *et al.* (2022) employed a comprehensive approach, utilizing both ^{31}P -NMR and LC-MS for phospholipid (PL) analysis and Gas chromatography for fatty acid composition. This investigation uncovered substantial variations in PL concentrations, composition, and fat globule sizes among milk samples from different animal groups. Notably, the observation that human milk demonstrated a fat globule size similar to that of bovine milk stands out as particularly significant. These cumulative findings propose a promising avenue for the application of Nuclear magnetic resonance (NMR) analysis in refining infant formulas to closely emulate

human milk, thereby enhancing their nutritional quality.

Notably, goat milk has been identified as particularly nutrient-dense compared to bovine milk. Researchers have conducted investigations into the metabolites of goat milk using GC-MS, exploring various bioactive compounds with reported medical benefits and hypoallergenic properties. Studies highlight the potential health advantages of these bioactive mixtures. In the realm of cheese manufacturing, GC-MS-based metabolomic analysis has identified creatinine and 4-hydroxyphenyl acetic acid as potential biomarker metabolites present in goat milk, demonstrating their utility in the cheese-making process (Hjerpsted *et al.*, 2014; Zheng *et al.*, 2015). The application of GC-MS-based metabolomics has further been instrumental in determining the composition of goat milk. Chilliard *et al.* (2006) and Avondo *et al.* (2015) utilized metabolomic examination of milk components such as lactose, fat, protein, and urea to validate the relationship between genotype and goat milk production, underscoring the significant role of genotype in determining milk composition.

Metabolomics to analyse effects of processing: The metabolomic analysis is best performed on milk and milk products. Mechanical boundaries are significant in the metabolomic examination of endless milk items. Rennet coagulation and syneresis of curd, which are related to creating cheese, are significant milk processing stages at dairies. According to the glycosylation state of the individual casein molecules, rennet coagulation is an enzymatic breakdown of casein that results in para-casein and hydrophilic casein-macro peptide (CMP) or glycomacropeptide (GMP). Breed, protein composition, species and hereditary variations of proteins influence the coagulation milk properties (Bittante *et al.*, 2012). Sundekilde *et al.* (2013) reported that the metabolite profile of cow milk influences its coagulation properties. These authors further believed that choline was positively correlated with the well-coagulation ability of milk. While, carnitine, citrate and lactose are significant metabolites that negatively correlated with milk's coagulation properties.

Heat stability is likewise another mechanical boundary managed by milk metabolites. Several chemical changes take place during heat treatment (Zabbia *et al.*, 2012). The heat stability of milk was investigated by analysing phosphorus-containing compounds in UHT-treated milk by ^{31}P NMR

spectroscopy (Belloque *et al.*, 2001). The casein content following sanitization or UHT treatment is not entirely settled by NMR spectroscopy (Belloque and Ramos, 1999). Numerous phosphorus-containing compounds in buffalo milk analysed by ^{31}P NMR spectroscopy, such as N-acetylglucosamine-1-phosphate, galactose-1-phosphate, glucose-6-phosphate and glycerol-1-phosphate (Andreotti *et al.*, 2000; Andreotti *et al.*, 2006; MacKenzie *et al.*, 2009). The authors also reported that cow and buffalo milk contained the same classes of phosphorylated compounds with some variability between the two. Jansson *et al.* (2014) identified some amino acid formation during the storage of UHT-treated milk by using NMR-based metabolomics. It has additionally been shown that UHT handling mixed with lactose hydrolysis makes lactose-reduced milk susceptible to substance changes and proteolysis during capacity (Jansson *et al.*, 2014). ^1H NMR metabolomics information upheld by customary biochemical examination is a valuable device to survey metabolic changes coming about because of the communication of pre-and probiotics in cheddar (Rodrigues *et al.*, 2011). Different authentic food intake biomarkers that are necessary for the human diet are identified by GC-MS-based metabolomics. Short-chain unsaturated fats and cholesteryl esters are potential biomarkers that can be related to GC-MS-based examination (Jenkins *et al.*, 2017).

Metabolomics for quality assessment and authentication: In recent years, there has been an increasing emphasis on ensuring the quality and authenticity of food products, including milk and its derivatives. To address this challenge, metabolomics has emerged as a powerful tool for comprehensive quality assessment and authentication of milk and milk products. Metabolomic examination assumes a significant part in deciding the nourishing nature of endless milk items.

Jia *et al.* (2020) introduced an untargeted MS-metabolomics approach to prevent fraud in dairy industries. They observed differences in molecule profiles of raw milk from cow, goat, and buffalo milk for discriminate analysis. Cow milk contained β -carotene, while water buffalo milk had ergocalciferol. Goat milk had higher decanoic acid and octanoic acid content than cow and water buffalo milk. This approach enables the comprehensive identification of unknown compounds and helps prevent fraud in the dairy industry.

Drug residues and antibiotics are found in animal

tissue and products, causing side effects in humans. The illegal use of veterinary drugs has become a major concern. New detecting methods, such as metabolomics, have been developed to control the illegitimate use of unknown drugs or mixtures of veterinary drug residues in cattle. Liquid chromatography coupled with Electrospray ionization and Tandem mass spectrometry (LC-ESI-MS/MS) has been used to quantify and identify 115 veterinary drug and pharmaceutical residues in animal-derived foods (Dasenaki and Thomaidis, 2015). Tandem capillary electrophoresis (CE) and Mass spectrometry (MS) have been applied to quantify fluoroquinolones in bovine milk, specifically ciprofloxacin, norfloxacin, and ofloxacin, through the coupling of CE with off-line CE-MALDI-TOF spectrometry (Springer *et al.*, 2015). Similarly, a targeted approach utilizing two-dimensional LC has proven successful in analyzing the presence of 20 antibiotic residues, spanning β -lactams, aminoglycosides, amphenicols, quinolones, and sulfonamides, in various dairy products, including powdered milk, commercial milk, and raw milk (Wang *et al.*, 2017).

NMR has been extensively used to study cheeses, focusing on factors such as geographic origin, molecular water mobility, and water retention capacity. In a study by Marseglia *et al.* (2013), the ripening and geographic discrimination of Parmigiano Reggiano Italian cheese were analysed using ^1H NMR. The study found that the ripening and production environment may influence the amino acid content, with high levels of leucine and iso-leucine at the initial ripening stages and high levels of threonine at the late ripening stages. The cheeses' characteristic flavour and geographic origin were also determined.

Using ^1H TD-NMR technology, researchers could differentiate cheese produced by small farms from those produced by dairy industries. The results showed that the fatty acid composition of the cheeses produced by small farms had higher levels of unsaturated fatty acids (oleic, linoleic, linolenic, and conjugated linoleic acids) compared to the industrially produced cheese (Segato *et al.*, 2019). This may be related to the cow's diet, which includes high amounts of food concentrates, compared to bovines from farms, which eat natural pastures at high altitudes. This highlights the importance of cattle nutrition in affecting milk quality and dairy products like cheese.

Coimbra *et al.* (2020) evaluated the effectiveness of ^1H TD-NMR in detecting formaldehyde addition in milk samples. The study found a direct correlation between relaxation times and formaldehyde

concentration, as higher concentrations increased water and fat displacement in raw milk. The NMR analysis revealed an increase in transversal relaxation time (T_2) due to changes in water and fat mobility during coagulation. This suggests that NMR may be an effective alternative for the dairy industry in detecting adulteration of raw milk by formaldehyde.

Within the realm of microbial metabolomics, pivotal strides have been made to unravel the intricate dynamics influenced by environmental factors in complex biological systems, as exemplified in notable studies by Cevallos-Cevallos *et al.* (2011) and Chen *et al.* (2022). Employing Gas chromatography-mass spectrometry (GC-MS), researchers have adeptly characterized prevalent food-borne pathogens such as *Escherichia coli*, *Listeria monocytogenes*, and *Salmonella enterica*. This approach not only discerns potential pathogen-specific biomarkers but also facilitates a rapid and accurate differentiation between uncontaminated and contaminated food samples (Jadhav *et al.*, 2019). The various microbial toxicants, encompassing amatoxins, lectins, phytotoxins, enterotoxins, mycotoxins, neurotoxins, and haemolysins, have been meticulously unveiled through metabolomics analysis, shedding light on this diverse array of threats.

Conclusion

Metabolomics is a promising field in the dairy industry, offering insights into production and product development. It can improve quality control, authenticity assessment, nutritional profiling, herd management, disease detection, and sustainable farming practices. Technological advancements will refine analytical techniques, making metabolomics a cornerstone for innovation, sustainability, and quality assurance in the dairy sector. It also contributes to the early detection of metabolic disorders in dairy animals.

Conflict of interest: Authors have no conflict of interest in this study.

Author's contribution: PAM: Writing original draft; TH: Conceptualization, writing, review and editing; KA: Conceptualization, writing, review and editing; RG: Conceptualization, writing, review and editing; RS: Writing original draft.

Data availability statement: No specific research data was used for the review article and information is compiled from the available literature.

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