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Effect of plant-derived additives on bioactive properties of fermented dairy products : A review

K.K.B.S. Rajapaksha^{1*}, M.A.L.S.S. Munasinghe¹, H.N.N. Dilrukshi¹, W.A.D. V. Weerathilake¹

¹Department of Livestock and Avian Sciences, Faculty of Livestock, Fisheries and Nutrition, Wayamba University of Sri Lanka, Makandura, Gonawila (NWP), Sri Lanka

Abstract

Food bioactives are compounds that exert physiological benefits promoting health and preventing effects of diseases in the human body through their antioxidant, antibacterial, anticancer, antidiabetic and analgesic properties. Plant-derived bioactive owing to the growing researcher's interest due to their abundance, low cost and wider acceptability. This growth is fuelled by technological innovations through the development of new products by fortifying plant-based sources. Fermented dairy products are of great importance due to their special characteristics, are an excellent matrix for the incorporation of ingredients and benefits on the hosts intestine and microbiome, immunomodulation and anti-allergenic effects. This review has extensively assessed the recent knowledge on the quantitative and qualitative enhancement of the nutritional value of fermented milk products by the enrichment of plant-based bioactive compounds. Fermented dairy products including yoghurt, curd, kefir, sour cream, cheese, buttermilk and drinking yoghurt have popularly fortified with different fruits, vegetables, herbs, spices, cereals and nuts. The findings of this review demonstrated that the bioactivity of fermented dairy products has increased with the incorporation of fruits, since they contain the majority of bioactive compounds such as carotenoids, polyphenols, dietary fibre and fatty acids. Fermented dairy products supplemented with herbs are typically high in polyphenols, fatty acids and carotenoids. It was also shown that spices enriched dairy products had more polyphenols, carotenoids and dietary fibre than unfortified dairy products. Nuts and oil crops, such as walnuts and hazelnuts have enhanced the fatty acid content of yoghurts. Taken together, these findings suggest that the role of bioactive compounds in promoting antioxidant, antimicrobial, antihyperglycemic, anti-inflammatory and antiradical effects. In future, it would be interesting to assess more studies on the synergetic effect between the natural bioactive compounds and fermented dairy products to enhance growing demand and potential health benefits.

Key Words: *Fermented Dairy Products*¹, *Health Benefits*², *Nutritional Value*³, *Plant-based Bioactives*⁴

Introduction

The consumption of fermented dairy products has seen a significant and steady increase over recent years, largely due to their enhanced digestibility, extended shelf life, improved nutritional and organoleptic properties, and the reduction of antinutritional compounds generated during fermentation. Certain strains of lactic acid bacteria are capable of metabolizing antinutritive components such as lactose and galactose, thereby offering protection against lactose intolerance and galactose accumulation (García-Burgos et al., 2020). Popular fermented dairy products include yogurt, curd, kefir, sour cream, cheese, buttermilk, and drinkable yogurt, all of which have been widely studied for their functional and health-promoting properties.

During fermentation, a variety of bioactive compounds such as vitamins, gamma-aminobutyric acid (GABA), bioactive peptides, bacteriocins, enzymes, conjugated linoleic acid (CLA), and exopolysaccharides are produced. These compounds have demonstrated potential roles in cancer prevention, blood pressure regulation, and type 2 diabetes management (Linares et al., 2017; Lee et al., 2020). Additionally, the increasing demand for fermented dairy products among individuals with lactose intolerance further highlights their functional appeal.

Despite the nutritional richness of fermented dairy products, some micronutrients such as zinc may be present in insufficient quantities (Jalal et al., 2016), and processing methods like heat treatment can degrade heat-sensitive nutrients. Moreover, fermentation may sometimes reduce antioxidant activity in products like yogurt (Dabija et al., 2018). As a result, fortification becomes essential to maintaining or enhancing the overall nutritional profile of these products.

The growing interest in functional foods—defined as those that provide health benefits beyond basic nutrition when consumed as part of a regular diet (Products & Foods, 1998)—has driven innovation in the dairy sector. With rising concerns over the negative effects of synthetic additives on the nervous and gastrointestinal systems, and their potential carcinogenic risks (Alenisan et al., 2017), researchers have turned their attention to natural plant-based additives rich in bioactive compounds.



Bioactive compounds—non-essential nutritional molecules present in small amounts—offer multiple health benefits, including antioxidant, antimicrobial, antidiabetic, anticancer, and analgesic effects (Septembre-Malaterre et al., 2017). These include phytosterols, polyphenols, unsaturated fatty acids, dietary fiber, and pigments such as carotenoids, anthocyanins, chlorophylls, and lycopene, commonly found in herbs, spices, fruits, vegetables, and green tea. Incorporating these compounds into dairy products can enhance both nutritional quality and product shelf life, while contributing positively to consumer health.

To effectively enrich dairy products with these bioactive compounds, a range of extraction and encapsulation techniques are employed, such as Soxhlet extraction, ultrasound-assisted extraction, supercritical fluid extraction, spray drying, coacervation, and freeze drying (Martins et al., 2017). However, challenges such as high dosage requirements, low bioavailability, unclear mechanisms of action, and limited availability of raw materials may arise. Advanced approaches involving biotechnology, nanotechnology, active packaging, and sustainable sourcing have been proposed to overcome these issues (Prakash et al., 2017).

Globally, the nutritional and therapeutic potential of natural sources has stimulated extensive research in this domain. Studies have shown that fortification with plant-based antioxidants can effectively reduce oxidation in dairy products with fewer adverse effects compared to synthetic additives (Alenisan et al., 2017). While essential oils have demonstrated positive antioxidant and antimicrobial properties, they may also affect sensory attributes negatively (Mishra et al., 2020). Similarly, the incorporation of grape extracts has improved antioxidant activity in dairy matrices but negatively impacted flavor (Kandylis et al., 2021).

Despite these advancements, research on the incorporation of tropical plant-based additives into fermented dairy products in Sri Lanka remains limited. The local application of such additives, which are rich in bioactive compounds, holds considerable potential for improving public health. Although antioxidant and antimicrobial properties have been well-documented, there is a notable lack of studies on the anti-tumor, anti-carcinogenic, anti-mutagenic, and immunomodulatory effects of these compounds when used in dairy product fortification (Prakash et al., 2017).

The primary aim of this review is to critically evaluate how natural additives rich in bioactive compounds can enhance the nutritional and functional quality of fermented dairy products, both quantitatively and qualitatively. Additionally, this review seeks to identify promising plant-based sources for fortification and to assess their impact on sensory, nutritional, and health-related attributes. A survey of Sri Lanka's dairy industry was also conducted to understand current practices and perceptions regarding the use of bioactive-rich natural additives in fermented dairy products. Ultimately, this article presents a comprehensive overview of fermented dairy products, the importance of fortification, the functional potential of plant-derived bioactive compounds, and the challenges and prospects in this emerging field.

Materials and Methods

A comprehensive literature review was conducted using the Web of Science database to gather relevant studies aligned with the research objectives. A total of 291 articles were initially selected based on specific keywords related to fermented dairy products, fortification, and bioactive compounds. To maintain relevance and consistency, articles published before 2016 and those written in languages other than English were excluded. Following a thorough screening process, 40 articles that met the inclusion criteria were selected for detailed review and analysis. The selected studies were assessed based on their contribution to understanding the nutritional, sensory, and health-related impacts of incorporating bioactive compound-rich natural additives into fermented dairy products.

Due to the extensive body of literature, the focus was narrowed to four major classes of bioactive compounds commonly studied in food fortification: polyphenols, carotenoids, dietary fiber, and polyunsaturated fatty acids. Similarly, among the wide range of fermented dairy products, cheese, yogurt, kefir, butter, and fermented milk were selected due to their global popularity and variation in formulation.

The selected articles were first qualitatively analyzed to understand the role and impact of each bioactive compound on nutritional enhancement, sensory attributes, and potential health benefits. Subsequently, the magnitude of observed effects was assessed and summarized, allowing for a more structured comparison of the outcomes across different studies.



Results and Discussion

Yogurt

Lactobacillus bulgaricus and *Streptococcus thermophilus* are the primary starter cultures used in the fermentation of cow's milk to produce yogurt. Yogurt is a rich source of minerals such as calcium, magnesium, phosphorus, and zinc, along with essential vitamins including B1, B2, B12, and D. It offers several health benefits, including enhanced lactose digestion, improved bioactive compound profiles, and increased bone mineral uptake (Gómez-Gallego et al., 2018).

Yogurt is particularly beneficial for individuals with lactose intolerance due to the partial hydrolysis of lactose by bacterial β -galactosidase. Furthermore, it plays a preventive role in various health conditions, including cancer, hypertension, and diarrhea, and supports gut microbiota balance. The unique flavor of yogurt is attributed to the volatile compounds produced by fermentative bacteria.

Yogurt can be categorized based on fat content, physical structure, and flavoring (Chandan, 2017):

Fat content: High-fat, reduced-fat, low-fat yogurt

Physical structure: Set yogurt, stirred yogurt

Flavorings: Plain yogurt, flavored yogurt

Figure 1. Classification of yogurts (Source: Chandan, 2017)

The general processing steps for yogurt production include standardization and fortification, homogenization (65°C, 1.8×10⁴ kPa), heat treatment (85°C for 30 minutes), cooling to incubation temperature (42°C), and inoculation with the starter culture.

Figure 2. Flow diagram of yogurt processing (Source: Chandan, 2017)

The current study affirms that yogurt can be effectively enriched with bioactive compounds through natural additives (Jaster et al., 2018; Šeregelj et al., 2019, 2021a).

Cheese

Cheese is a versatile dairy product available in cooked and uncooked varieties. Over time, its nutritional value, texture, and flavor have improved. Cheese offers numerous health benefits, such as supporting bone and dental health, regulating blood pressure and cholesterol, and maintaining vascular and gut health (Martins et al., 2017).

Cheese can be made from various milk sources including cow, goat, sheep, and buffalo milk. Based on processing parameters, cheese is classified into several types (Hammam & Ahmed, 2019):

Processing method: Cheddar, cream cheese, cottage cheese

Composition: Salted, unsalted, high-fat, low-fat cheese

Maturation method: Acid-ripened, enzymatic ripened

Starter culture: Mold-ripened, bacteria-ripened cheese

The general processing steps for cheese involve standardization and pasteurization, inoculation with cultures, renneting, curd cutting, cooking (37°C–42°C for 40 minutes), salting, forming, pressing, and aging.

This study demonstrates that cheese can be successfully fortified with natural additives rich in bioactive compounds (Balabanova et al., 2020; Ghendov-Moşanu et al., 2020; Masmoudi et al., 2020).

Kefir

Kefir, an emerging functional dairy product, exists in two main forms: milk kefir and water kefir (Guzel-Seydim et al., 2021). It contains high levels of organic acids, sugars, esters, and alcohols, contributing to its distinct flavor and texture.

Kefir is produced by fermenting milk with kefir grains or lyophilized cultures that primarily consist of lactic acid bacteria and yeast. Nutritionally, kefir is abundant in B-complex vitamins (B1, B2, B6, B12), essential amino acids (e.g., phenylalanine, tyrosine, leucine, glycine), and minerals such as magnesium, potassium, and calcium (Vicessuto & de Castro, 2020a).

Due to its probiotic, prebiotic, antibacterial, anti-inflammatory, and antioxidant activities, kefir offers protective effects against obesity, hypercholesterolemia, and cancer while promoting longevity. The study confirms that kefir can be effectively enriched with natural additives containing bioactive compounds (Vicessuto & de Castro, 2020a, 2020b).

Butter

Butter typically contains 80–83% fat and up to 400 different fatty acids. It is widely used both as a spread and a cooking fat. Butter is produced by churning milk or cream, which disrupts fat globules and forms a water-in-oil emulsion, separating butter granules from buttermilk.

While cow milk is most commonly used, butter can also be derived from sheep, goat, and buffalo milk (Mehdizadeh et al., 2019). Chemically, butter is rich in fat, protein, milk sugars, and water. However, it is highly susceptible to spoilage through lipid oxidation and microbial contamination, requiring stringent storage conditions (El-Hadad & Tikhomirova, 2018).



This study supports that butter can also be enriched successfully with bioactive compounds via natural additives (Mehdizadeh et al., 2019; Vidanagamage et al., 2016).

Importance of Fortification of Fermented Dairy Products

Fermented dairy products are nutrient-dense, supplying protein, calcium, vitamins (especially B12 and riboflavin), and essential minerals. Individuals with lactose intolerance can tolerate fermented dairy due to bacterial conversion of lactose into lactic acid. Additionally, these products exhibit antibacterial, antitumor, and immunomodulatory properties (Savaiano & Levitt, 1984).

Fortification improves the nutritional profile of fermented dairy products, making them more functional and health-promoting. Moreover, the matrix properties of dairy products (texture, composition, and pH) play a crucial role in the efficiency and stability of added bioactive compounds.

Bioactive Compounds in Natural Sources

Bioactive compounds are secondary plant metabolites that contribute to a plant's adaptability and survival, though not directly involved in growth or reproduction. These compounds include phenolics, alkaloids, terpenes, and phytosterols (Galanakis & Drago, 2022).

Phenolic compounds, in particular, are further classified into flavonoids and non-flavonoids (Kamiloglu et al., 2021).

Figure 5. Classification of bioactive compounds (Source: Khezerlou & Jafari, 2020)

Extraction methods for these compounds include conventional techniques like Soxhlet and hydrodistillation, and advanced methods such as ultrasound-assisted extraction (UAE), microwave-assisted extraction (MAE), supercritical fluid extraction (SFE), pressurized liquid extraction (PLE), and pulsed electric field extraction (PEF).

Phenolic Compounds: Abundance, Bioactivity, and Application in Functional Foods

Phenolic compounds represent the most abundant class of secondary metabolites in plants, characterized by the presence of one or more hydroxyl groups attached to aromatic rings (Ayad & Akkal, 2019). These compounds are widely distributed among various plant species and are classified into several subgroups, including flavonoids, phenolic acids, lignans, stilbenes, and tannins.

Phenolics are known for their diverse biological activities. One of their notable functions is their anti-inflammatory property, which contributes significantly to the prevention of cardiovascular diseases. This effect is primarily exerted through the inhibition of platelet aggregation and modulation of smooth muscle cell activity (Guasch-Ferré et al., 2017). Furthermore, phenolics exhibit antidiabetic effects by enhancing glucose uptake in peripheral tissues and reducing insulin resistance through improved insulin sensitivity (LS & NJA, 2016). Their anti-obesity potential is associated with the suppression of adipocyte proliferation, stimulation of lipolysis, appetite regulation, and reduction of inflammation in adipose tissue (LS & NJA, 2016).

The anticancer potential of phenolic compounds is also well documented. They exert protective effects by scavenging free radicals, inducing detoxifying enzymes involved in xenobiotic metabolism, regulating gene expression, and modulating several signaling pathways involved in DNA repair, apoptosis, and cell invasion (Lutz et al., 2019; Yen et al., 2020).

Recent research has explored the functional properties of phenolics in food systems. Masmoudi et al. (2020) reported that the addition of 30% strawberry pulp to a food matrix resulted in a significant increase in anthocyanin content by 2.99 mg/100 g, along with enhanced antioxidant activity measured by DPPH (9.19 mg GAE/mL) and ABTS (0.38 mg GAE/mL) assays. Although a decrease in anthocyanin content was observed during storage, the antioxidant activity remained relatively stable.

In a comparative study, Muniandy (2016) found that yoghurt enriched with green tea exhibited the highest total phenolic content (526.34 mg GAE/mL) compared to those enriched with white tea (394.12 mg GAE/mL) and black tea (294.67 mg GAE/mL), likely due to the higher catechin and epicatechin content in green tea. However, storage over two weeks led to a significant decline in phenolic content across all samples. Antioxidant activity, evaluated by DPPH RSA and FRAP, was highest in green tea yoghurt, while black tea yoghurt showed superior antioxidant potential in the FIC assay. These variations may reflect the differences in primary and secondary antioxidant capacity measured by these methods.

Josipović et al. (2015) demonstrated that the addition of dried dill significantly increased the phenolic content of cheese to 37.8 mg GAE/100 g, surpassing that of cheese fortified with parsley, pepper, garlic, or rosemary. However, this increase in phenolic content did not correspond with a higher antioxidant effect. The discrepancy was attributed to several factors, including pH, temperature, phenolic structure, amino acid composition, and potential interactions between phenolics and proteins. Additionally, the capacity of certain reagents to react with non-phenolic compounds may influence the accuracy of antioxidant measurements.

Vidanagamage et al. (2016) investigated the incorporation of cinnamon extract into butter and observed an increase in total phenolic content to 135.62 µg GAE/mg and antioxidant activity to 38.73%, highlighting the potential of spice extracts in enhancing the functional properties of dairy products.



Kabakcı et al. (2020) examined kefir enriched with anthocyanin-rich fruit juices and found that the addition of 25% black mulberry juice led to the highest anthocyanin content (404.5 mg/L) compared to pomegranate, strawberry, and black carrot juices. However, anthocyanin degradation and co-pigmentation during storage led to a significant reduction (32.1%) in anthocyanin content. The antioxidant effect was associated with the anthocyanin content of black mulberry, although no notable antimicrobial activity was detected.

Contradictory findings were reported by Kiros et al. (2016), who found no increase in phenolic content in carrot juice-enriched yoghurt compared to the control, contrary to results by Pandey et al. (2021). In fact, a slight decrease from 37 mg GAE/kg in the control to 35.75 mg GAE/kg in the fortified sample was recorded. Possible explanations include differences in carrot varieties, agronomic practices, storage conditions, and processing techniques used prior to analysis.

Carotenoids

Carotenoids are lipid-soluble secondary metabolites widely distributed in nature, especially in plants, where they play essential roles in photosynthesis and photoprotection. From a nutritional perspective, carotenoids are recognized for their health-promoting properties, including provitamin A activity, antioxidant potential, and disease-preventive roles (Karpinski et al., 2022).

Structurally, carotenoids are composed of isoprene units forming C40 terpenoids, and they are typically classified into two main groups: carotenes (hydrocarbon carotenoids, such as α -carotene, β -carotene, and lycopene) and xanthophylls (oxygenated carotenoids, including lutein and bixin). Carotenes are characterized by non-polar structures, while xanthophylls contain polar groups, making them more reactive to oxidation. Among these, lycopene has the simplest linear structure, whereas α - and β -carotenes possess cyclic ends. The differentiation between α - and β -carotene arises from the position of the double bond in the cyclic group (Karpinski et al., 2022). Carotenoids play crucial roles in quenching singlet oxygen and scavenging reactive oxygen species (ROS), attributed to the conjugated double bonds in their polyene chain. This ability to deactivate ROS stems from mechanisms such as triplet-triplet energy transfer, electron transfer, hydrogen atom donation, and radical adduct formation (Karpinski et al., 2022). Furthermore, carotenoids' antioxidant activity is directly proportional to the number of conjugated double bonds in their structure (Zakynthinos & Varzakas, 2016).

In addition to antioxidant capacity, carotenoids exhibit anti-inflammatory and antimicrobial activities. They interact with the NF- κ B pathway, inhibiting its nuclear translocation and consequently reducing the expression of inflammatory cytokines such as interleukin-8 and prostaglandin E2 (Kaulmann & Bohn, 2014). Moreover, carotenoids have been reported to interfere with efflux pump activity in bacteria by disrupting ATP hydrolysis, altering membrane permeability, and increasing intracellular ROS production, thereby exerting antibacterial effects (Zakynthinos & Varzakas, 2016).

Carotenoids also contribute to cancer prevention by modulating signaling pathways associated with apoptosis, cell cycle arrest, and metastasis. Their chemopreventive roles include inhibition of tumor cell proliferation and enhancement of programmed cell death (Niranjana et al., 2015).

Numerous studies have explored the fortification of fermented dairy products with carotenoid-rich sources to enhance their functional properties. Kiros et al. (2016) reported that yogurt fortified with carrot or carrot oil demonstrated significantly higher carotenoid content and antioxidant activity compared to the control. Specifically, the addition of carrot oil resulted in a carotenoid concentration of 10.26 mg/kg in fresh yogurt. Similarly, a study by Balabanova et al. (2020) evaluated the impact of incorporating different pepper varieties into yogurt. Among them, yogurt enriched with *Kurtovska Kapia* showed the highest carotenoid content (570.02 μ g/100g), exceeding the levels found with other varieties such as *Doux Marconi Geonet* and *Bulgarian Ratunt*.

These findings highlight the potential of carotenoid fortification in enhancing the health-promoting properties of dairy products, making them valuable functional foods in modern diets.

Dietary Fiber

Dietary fiber refers to the indigestible portion of plant-derived carbohydrates, primarily in the form of polysaccharides, that escapes enzymatic digestion in the human small intestine. These fibers are broadly classified based on their water solubility into two major types: soluble and insoluble fibers. Soluble dietary fibers include pectin, gums, mucilage, fructans, and specific types of resistant starches, which are commonly found in fruits, vegetables, oats, and barley. In contrast, insoluble fibers such as lignin, cellulose, and hemicellulose are mainly sourced from whole grains, nuts, seeds, and vegetables (Soliman, 2019).

The inclusion of dietary fiber in daily intake has been associated with a reduction in the risk and severity of a range of health disorders, including constipation, diverticulosis, colorectal cancer, type 2 diabetes, obesity, cardiovascular diseases, and gallstones. Moreover, increased fiber intake has been linked to a reduction in serum cholesterol and blood pressure levels (Li & Komarek, 2017).

An increasing body of research has focused on the enrichment of dairy products with plant-derived dietary fiber sources. Issar, Sharma, and Gupta (2017) reported that the incorporation of 5% apple pomace fiber into acidophilus yogurt resulted in an acceptable sensory profile and enhanced fiber content, making it a suitable approach for



developing fiber-enriched fermented dairy products. Similarly, Kurt and Atalar (2018) demonstrated that the addition of quince seed-derived dietary fibers significantly increased the viscosity of yogurt, from 0.266 to 0.317 Pa·s, potentially improving the textural attributes and functional value of the product.

Fatty Acids

Fatty acids (FAs) are fundamental lipid components classified based on their saturation level. Saturated fatty acids (SFAs), such as palmitic acid (C16:0), stearic acid (C18:0), myristic acid (C14:0), and lauric acid (C12:0), are characterized by the absence of double bonds in their hydrocarbon chains. Unsaturated fatty acids (UFAs), which include monounsaturated (MUFAs) and polyunsaturated fatty acids (PUFAs), are distinguished by the presence of one or more double bonds (Karaguzel et al., 2019). Notably, oleic acid (C18:1), linoleic acid (LA, C18:2, omega-6), and alpha-linolenic acid (ALA, C18:3, omega-3) are the predominant UFAs found in plant sources.

PUFAs, particularly omega-3 fatty acids like ALA, eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA), are recognized for their beneficial effects in preventing cardiovascular diseases, modulating inflammation, and supporting neurological development (Dal Bello et al., 2017). Conversely, a disproportionate intake of omega-6 PUFAs, especially in the absence of adequate omega-3s, has been associated with adverse health outcomes (Shrestha et al., 2020).

In recent years, extensive research has focused on the fortification of dairy products with plant-derived oils rich in essential fatty acids. Ozturkoglu-Budak et al. (2016) observed that yogurt enriched with walnuts exhibited a significant increase in total fatty acid content (69.54 g/100 g) compared to variants fortified with hazelnuts, almonds, or pistachios. This enhancement was attributed to the high EPA and DHA content in walnuts, which also contributed to lower stiffness development during storage.

Similarly, Van Nieuwenhove et al. (2019a) demonstrated that yogurt fortified with pomegranate seed powder exhibited a 1.8-fold increase in conjugated linolenic acid (CLnA) compared to yogurt fortified with Jacaranda seed. The addition of these plant powders did not adversely affect the pH or sensory properties of the yogurts, even after 28 days of storage (Van Nieuwenhove et al., 2019b).

Dal Bello et al. (2017) reported that cheese samples fortified with blackcurrant oil presented the highest omega-6 PUFA levels (12.21 mg/g) on day one, surpassing samples fortified with raspberry, flaxseed, camelina sativa, and echium oils. The greatest omega-3 content (4.43 mg/g) was recorded in cheeses fortified with camelina oil. Interestingly, flaxseed oil-fortified cheeses exhibited a significant decrease in omega-3 content after three days of ripening. No significant differences were found in protein, moisture, ash content, or pH among control and fortified cheeses. Among the oil treatments, the combinations of flaxseed and raspberry oils were found to be the most favorable in terms of fatty acid composition and consumer acceptance.

Table 01 - Natural sources rich in bioactive compounds

Natural Source	Examples	Phenolic Compound	Reference
Herbs	Red ginseng	Polyphenols	(H. Park et al., 2018)
	Sea buckthorn	Carotenoids	(Ghendov-Moşanu et al., 2020)
Spices	Turmeric	Carotenoids	Hasneen et al., 2020)
	Ginger	Phenolics	(Ahmed et al., 2021a)
	Dried curry leaves	Phenolics	(Weragama et al., 2021)
	Pepper	Carotenoids	(Balabanova et al., 2020)
	Cinnamon	Phenolics	(Vidanagamage et al., 2016)
	Strawberry	Phenolics	(Jaster et al., 2018)
	Passion	Phenolics	(Ning et al., 2021)
	Pomegranate	Phenolics	(Van Nieuwenhove et al., 2019b)
	Mango	Carotenoids	(Vicenssuto & de Castro, 2020a)
Vegetables	Carrot	Carotenoids	(Kiros et al., 2016)
	Spinach	Phenolics	(Göksel Saraç & Dogan, 2016)
Nuts and oil crops	Walnut	Fatty acids	(Ozturkoglu-Budak et al., 2016)
	Hazelnut	Fatty acids	(Ozturkoglu-Budak et al., 2016)
	Almond	Fatty acids	
Cereals	Black rice	Phenolics	(Khalil & Elkot, 2020)
	Wheat	Phenolics	(Çetinkaya & Öz, 2020)

Bioactive Compounds in Fermented Dairy Products

Fermented dairy products are rich sources of bioactive compounds, primarily produced through the metabolic activity of microorganisms such as lactic acid bacteria (LAB), yeasts, and molds. These microorganisms contribute to the synthesis of several functional metabolites, including exopolysaccharides (EPS), conjugated linoleic acid



(CLA), bioactive peptides, gamma-aminobutyric acid (GABA), galactooligosaccharides (GOS), and essential vitamins (Linares et al., 2017).

The vitamin profile of fermented dairy products is notably enhanced, especially in riboflavin and vitamin B12, which are synthesized during fermentation by specific strains of LAB. The consumption of such fortified products may contribute to reducing vitamin deficiencies in human populations (Savaiano & Levitt, 1984). Moreover, GABA is synthesized through the decarboxylation of glutamic acid by LAB and plays a vital role in human neurological function, contributing to the regulation of neurotransmission and stress responses (Santos-Espinosa et al., 2020).

Exopolysaccharides, also produced by starter cultures, are recognized for their dual functionality: enhancing textural attributes of fermented dairy products and promoting gut health by acting as prebiotic-like agents (Santos-Espinosa et al., 2020). In addition, bioactive peptides, typically composed of 2 to 20 amino acids, are generated via the proteolytic activities of microbial enzymes during fermentation. These peptides exert multiple physiological benefits, including antihypertensive, antioxidant, immunomodulatory, and antimicrobial effects, making them crucial targets in functional food research (Y. W. Park & Nam, 2015).

CLA, another key metabolite, exhibits strong antioxidant activity and is implicated in reducing the risk of chronic conditions such as cardiovascular disease, obesity, and certain cancers. Its inclusion in fermented dairy further enhances the health-promoting properties of these products, thereby supporting their role in maintaining a healthy lifestyle (Y. W. Park & Nam, 2015).

Collectively, these microbial-derived bioactive compounds not only improve the nutritional and functional quality of fermented dairy products but also underscore their potential in the prevention and management of various health disorders. This has spurred considerable scientific interest in optimizing fermentation conditions and microbial strains to enhance the bioavailability and efficacy of these compounds in functional dairy foods.

Natural sources of bioactive compounds used in fermented dairy products.

Health effect of fortification of fermented dairy products with bioactive rich natural compounds

Table 02a - quantitative evaluation of the effects of the bioactive compounds.

Dairy product	Natural Additive	Major bioactive ingredients	Experimental findings	Reference
Set yoghurt	Passion fruit juice (PFJ)	polyphenols	Total phenolic content (μg GAE/g) of set yoghurt increased from 1.69 to 11.4 with the addition of 2.5% PFJ. The antioxidant effect increased from 2.75% to 35.8% PFJ. (DPPH)	(Ning et al., 2021)
yoghurt	Red ginseng extract(RGE)	Polyphenols (ginseng saponins) Flavonoids	Total phenolic content (mg of GAE/100 g) and total flavonoid content (mg of GAE/100 g) of yogurt increased from 8.1 to 41.1 and from 8.4 to 18.7 with the addition of 2% red ginseng extract respectively. The antioxidant effect ($\mu\text{g/mL}$) of yoghurt increased from 5.8 - 13.1 to 18.7 - 41.1 with the addition of 2% red ginseng extract. (DPPH RSA)	(Park et al., 2018)
Probiotic yogurt	Green, black and white tea	Polyphenols	The highest total phenolic content (mg GAE/ml) recorded in green tea yoghurt (526.34). There was a significant reduction in TPC of all tea enriched yoghurts during storage. ($p<0.05$) The antioxidant activity of plain yoghurt (3-7%) increased into 83-97% with the addition of tea extracts ($p<0.05$)	(Muniandy et al., 2016)
yoghurt	Pomegranate seed (PS) and jacaranda seed (JS)	Fatty acids (conjugated linolenic acid) (CLnAS)	CLnAS of PS (62-63mg/100g) increased by 1.8-fold compare to JS yogurt. Highest antioxidant activity recorded in PS (61.55%). There was significantly decrease in antioxidant effect during 28 days.	(Van Nieuwenhove et al., 2019)



Table 02b - quantitative evaluation of the effects of the bioactive compounds.

Yoghurt	Walnut Hazelnut Almond Pistachio	Vitamins(folic acids, tocopherols) Minerals (selenium) Fatty acids	Highest folic acid amount (29.29 µg/100g) and fatty acid amount (69.54%) recorded in walnut enriched yoghurt. Highest Selenium (2.65 µg/100g) and γ-tocopherol (1.94 mg/100g) recorded in pistachio enriched yoghurt. Highest α-tocopherol content recorded in hazelnut enriched yoghurt (4.87 mg/100g). Almond enriched yoghurt had high protein (5.91wt/wt%) and walnut enriched yoghurt had high fat content(6.10wt/wt%). Lowest count of streptococci and <i>L. bulgaricus</i> recorded in walnut enriched yogurt.	(Ozturkoglu-Budak et al., 2016)
Dahi (Indian yoghurt)	Flax seed oi microcapsules	Fatty acids(α-linolenic acid)	A- linolenic acid (ALA) increased up to 10.62% with addition of 2% micro encapsulated flaxseed oil powder. ALA reduced (8.40) continuously during 15 days.	(Goyal et al., 2016)
Yoghurt	Carrot waste extract (CW)	Carotenoids	The highest beta-carotene bleaching antioxidant capacity(9.36 µmolTE/180g), reducing power(3.11 µmolTE/180g) recorded with 5CW beads enriched yoghurt. Highest antihyperglycemic activity(13.91%) and anti-inflammatory activity(15.27%) recorded with 5CW beads enriched yoghurt.	(Šeregelj et al., 2021)
Yoghurt	Seaweeds varieties (<i>Aacophyllum nodosum</i> , <i>fucus vesiculosus</i>)	Phenolics	Seaweed enriched yoghurts didn't exhibit cellular antioxidant activity.	(O'Sullivan et al., 2016)
Soft cheese	Clove Thyme Ginger	Phenolics (thymol, eugenol)	The minimum inhibitory concentration of clove, thyme and ginger essential oils was 0.001% for <i>S. aureas</i> , <i>E.coli</i> , <i>Ent. faecalis</i> , <i>P.aeruginosa</i> , <i>P.fluorascens</i> , <i>C. albicans</i> , <i>A. parasiticus</i> .	
Cream cheese	Dried curry leaves (DCL)	Phenolics	The highest phenolic content (193.33 mg GAE/100g) and highest antioxidant effect (lowest value of 0.06 IC ₅₀ (mg/ml) recorded with 0.25% DCL addition. Moisture and total solid content didn't significantly affect with DCL addition. Highest FRAP value (0.17Mm Fe(II) was recorded with 0.25% DCL addition. Lowest fat level (33.01%) recorded with 0.25% DCL addition.	(Weragama et al., 2021)
Soft cheese	<i>Arbutus unedo</i> L. (AUL)	Phenolics Dietary fiber	Highest antioxidant effect recorded with 1g/L AUL extract and it decreased during 5days of storage period. (DPPH method) Fat and ash didn't significantly affect by AUL addition. Protein (46%) significantly increased by 1g/L AUL addition.	(Masmoudi et al., 2020)



Table 02b - quantitative evaluation of the effects of the bioactive compounds.

Processed cheese spread	Black rice powder(BRP)	Crude fiber phenolics	<p>Highest total phenolic content (44.23 mg GAE/g) recorded by 6% BRP addition and it was reduced (36.62 mg GAE/g) gradually during 90days storage.</p> <p>Highest antioxidant scavenging activity 64.82% recorded 6% BRP addition and it was reduced (54.97) gradually during 90days storage.</p> <p>Dry matter and far were not significantly differed due to BRP addition.</p> <p>Protein (9.42%) was reduced by 6% BRP addition.</p> <p>Noticeable antimicrobial effect didn't recorded with BRP addition.</p>	(Khalil & Elkot, 2020)
Labneh cheese	Pepper varieties Doux Marconi geonet, balgarian ratunt, kurtovska kapia, Doux Marconi geonet x kurtovska kapia, Doux	Phenolics Carotenoids Chlorophyll	<p>The highest carotenoids (570.02 µg/100g), chlorophylls (960.29 µg/100g), total phenols (34.24GAE/100g), total flavonoids (0.05mg QE/100g) and antioxidant effect (28.95 Mm TE/100g) recorded with addition of kurtovska kapia variety.</p> <p>A slight decrease in phenols and flavonoids observed during storage.</p> <p>A significant difference for moisture, fat, total protein didn't recorded with addition of pepper.</p>	(Balabanova et al., 2020)
Cream cheese	Sea buckthorn lipophilic extracts(SBLE)	Carotenoids polyphenols	<p>28.89% antiradical activity recorded with addition of 2.5% SBLE.</p> <p>11.95% fat level recorded with 2.5% SBLE addition.</p> <p>Total viable count(CFU/g)*2 reduced from 54 to 10 with 2.5% SBLE addition after 10 days storage.</p>	(Ghendov-Moşanu et al., 2020)
Fresh cheese	Flaxseed oil(FS) <i>Camelina sativa</i> oil(CAM) Raspberry(RAS) <i>Echium plantagineum</i> oil(EC) Blackcurrent oil(BC)	Fatty acids(omega-3 FA and omega-6 FA)	<p>Highest omega-6 FA (12.21mg/g) recorded with addition of BC oil.</p> <p>Highest omega-3 FA (4.43 mg/g) recorded with addition of CAM oil.</p> <p>Omega-3 FA (α-linolenic acid) in FS enriched cheese showed a significance difference during storage.(p<0.01)</p> <p>Protein, moisture and ash didn't significantly differ with vegetable oil addition.</p> <p>Fat content increased (23.00%w/w) with the addition of RAS oil.</p> <p>Antimicrobial effect didn't show a significant difference among cheese samples.</p>	(Dal Bello et al., 2017)
White cheese	Wheat germ (WG)	Fatty acids	<p>High amount of poly unsaturated fatty (PUFA)acids and saturated fatty acids (SFA) recorded with 2% WG addition.</p> <p>SFA and PUFA increased significantly during 30 days storage.(p<0.05)</p> <p>Highest percentage of fat (13.93), protein (18.09), dry matter(42.03) and salt(3.85) recorded with 2% WG addition.</p> <p>fat, protein, dry matter and salt content increased significantly during storage (p<0.05, p<0.01)</p>	(Çetinkaya & Öz, 2020)
Butter	Olive pomace (OP) Olive mill waste water (OMWW)	Phenolics	<p>Total mesophilic aerobic flora of butters enriched with 8mg of OMWW and 8mg of OP showed zero level.</p> <p>Antibacterial and antifungal effects showed with OMWW and OP addition for total coliforms, yeast and moulds and <i>Staphylococcus aureus</i></p>	(Mikdame et al., 2020)



Table 02c - quantitative evaluation of the effects of the bioactive compounds.

Butter	Cinnamon Extract(CE)	phenolics	Total phenolic content($\mu\text{g GAE/mg}$) of 135.62 and antioxidant activity of 38.73% recorded in cinnamon enriched butter. Crude fat (82.12%), protein (0.76%), moisture (14.65%), CHO (0.08%), ash (2.39%) recorded with CE addition. Yeast and moulds weren't in cinnamon enriched yoghurt throughout the storage. Highest antimicrobial activity recorded in cinnamon enriched butter. (total plate count of 1.63×10^2 CFU/g at 8 th week).	(Vidanagamage et al., 2016)
Fermented milk	Pomegranate peel extract(PPE)	Phenolics	Significantly high total phenolic content and antioxidant activity recorded in fermented milk with PPE addition. The concentration below the 2% w/v didn't show an antimicrobial effect against <i>L. acidophilus</i>	(Chan et al., 2018)
Fermented milk	Grape marc (GM) Olive pomace (OP)	Phenolics	Highest total phenolics($98.5 \mu\text{g GAE/g}$) level recorded with OP addition and it decreased by 28% during 50days storage.($p < 0.05$) Highest antiradical power (ARP) (72.75%) recorded with OP addition and longer storage wasn't affected significantly on quantity of ARP. <i>S. thermophilus</i> count decreased not (statistically significant) during 14 days and the count was similar to control sample after 14 days <i>L. acidophilus</i> remained constant.	(Aliakbarian et al., 2015)
Kefir	Black mulberry juice(BMJ) Pomegranate juice(PJ) Strawberry juice(SJ) Black carrot juice(BCJ)	Phenolics (Anthocyanin)	Highest anthocyanin content(404.5 mg/L) and stability(32.1 weeks) recorded with 25% of BMJ addition. Antioxidant effects increased with 25% (1.8-4.8 times) compare to the plain kefir. Highest with BCJ addition-6.39). Highest antioxidant activity($6.39 \text{ mM Trolox/ml}$) recorded with 25% BCJ addition. Most decreased lactobacillus count showed in PJ 10% and 25% and SJ 25%. Lowest Lactococcus count recorded in PJ 25%.	(Kabakcı et al., 2020)
Kefir	Mango peel(MP)	Phenolics	21.35% increased antioxidant activity recorded in mango peel enriches kefir compared to the control kefir. Antioxidant activity increased 113%($13.81 \mu\text{mol TE/g}$) compare the control by addition of MP.(DPPH RSA method) effect on antimicrobial effect against acidic bacteria and yeast.	(Vicenssuto & de Castro, 2020)

Commonly Found Effects of Bioactive Compounds

Antioxidant Effect

The antioxidant effect refers to the ability of a compound to mitigate or prevent the formation of oxidizable substances during metabolism, thereby minimizing damage to the body (Brainina et al., 2019). Oxidation is primarily driven by reactive oxygen species (ROS) such as superoxide ions, singlet oxygen, lipid peroxides, and hydroxyl radicals. Antioxidants, which are abundant in fruits and vegetables, play a key role in reducing oxidative stress caused by these ROS (Serafini & Peluso, 2016).

Bioactive compounds derived from natural sources act by converting ROS into non-radical species, disrupting autoxidative chain reactions, and lowering local oxygen concentrations (Hunyadi, 2019). Oxidative stress is a contributing factor to various chronic diseases including cancer, cardiovascular diseases, diabetes, age-related muscular degeneration, and ocular disorders.



In fermented dairy products, the DPPH Radical Scavenging Activity (RSA) method is commonly employed to assess antioxidant activity. DPPH, a purple-colored radical, becomes colorless upon receiving electrons from antioxidants. Other analytical methods used include ABTS, FRAP, and FIC assays. Natural antioxidants include phenolic compounds, carotenoids, and vitamins, which not only contribute to health benefits but also improve product shelf-life and quality (Wilson et al., 2017).

For example, Masmoudi et al. (2020) reported increased antioxidant activity in products supplemented with 30% spirulina powder (SP), with DPPH RSA and ABTS values reaching 9.19 mg GAE/mL and 0.38 mg GAE/mL, respectively. Muniandy (2016) found that green tea-enriched yogurt showed the highest antioxidant activity according to DPPH RSA and FRAP methods, while black tea-enriched yogurt performed best in the FIC method. This suggests that DPPH RSA and FRAP assess primary antioxidant activity, while FIC evaluates secondary antioxidant activity. However, antioxidant activity tends to decline during storage due to phenolic degradation.

Antimicrobial Effect

Microorganisms in food can alter its color, texture, odor, flavor, and safety. Bioactive compounds, especially phenolics, possess antimicrobial properties against both Gram-negative and Gram-positive bacteria due to the presence of functional groups like hydroxyls (Khameneh et al., 2019). These compounds are found naturally in herbs, spices, fruits, vegetables, seeds, and leaves.

Phenolic compounds such as eugenol, thymol, carvacrol, vanillin, allicin, cinnamic aldehydes, and allyl isothiocyanate contribute significantly to the antimicrobial properties of herbs and spices (Ghendov-Moşanu et al., 2020; Ozturkoglu-Budak et al., 2016). Fatty acids and dietary fibers also play supporting roles (Khalil & Elkot, 2020; Mehdizadeh et al., 2019).

The mechanisms of antimicrobial activity include membrane disruption, pH reduction, and enzymatic inhibition. Microbial testing often targets yeasts, molds, coliforms, *Streptococcus* species, and psychrotrophs (Enab et al., 2012). A study by Ozturkoglu-Budak et al. (2016) found a positive correlation between plant-based fortification and antioxidant activity. Additionally, walnut-enriched yogurt showed the lowest counts of *Streptococcus* and *L. bulgaricus*.

Anti-Hyperglycemic Properties

Diabetes is one of the most prevalent non-communicable diseases globally. Bioactive compounds from natural sources like fruits, vegetables, herbs, and grains can lower blood glucose levels (Vinayagam et al., 2016). Phenolics are particularly effective, as they reduce insulin resistance, promote glucose uptake, increase glycogen synthesis, and inhibit carbohydrate-digesting enzymes such as α -glucosidase and α -amylase.

The efficacy of these compounds is often evaluated using α -glucosidase inhibition assays. Bioactives affect multiple organs, including the liver, pancreas, intestine, muscle, and adipose tissue. Šeregelj et al. (2021b) demonstrated that plant-based fortification correlates positively with anti-hyperglycemic activity. Kiros et al. (2016) found improved glycemic control in carrot juice-enriched products.

Anticarcinogenic Effect

Dietary habits play a significant role in the prevention of chronic illnesses, including various types of cancer. Epidemiological studies have linked high fruit, vegetable, and grain intake with reduced risks of heart disease, cancer, diabetes, Alzheimer's, and age-related decline (Subramaniam et al., 2019). The National Cancer Institute has recognized approximately 35 plant-based foods with potential anti-cancer properties, including garlic, soybeans, ginger, licorice root, and cruciferous vegetables such as broccoli and cabbage.

Oxidative stress, which damages macromolecules like DNA, proteins, and lipids, contributes to carcinogenesis and cardiovascular disease. Antioxidants, particularly phenolics and carotenoids from plants, mitigate this damage (Chandra et al., 2012). Rosemary, due to its phenolic hydroxyl groups, is a strong candidate for chemoprevention. Its bioactive compounds exhibit antimicrobial, antiviral, anti-inflammatory, and anticarcinogenic properties (Sereiti et al., 1999; Aherne et al., 2007; Moore et al., 2016).

Limitations

Extraction Method

Traditional extraction methods such as hydroalcoholic distillation, Soxhlet extraction, and maceration are commonly used to extract bioactive compounds. These methods often require high solvent volumes and long extraction times. In hydro distillation, high temperatures can degrade heat-sensitive compounds, potentially leading to inaccurate estimations (Ahmed et al., 2021b; Qiu et al., 2021; Ribeiro et al., 2016).

Soxhlet extraction, developed by Franz Rizz Von Soxhlet, uses solvents to extract oils and other bioactives but also exposes compounds to heat, risking degradation (Sasidharan et al., 2011). Maceration, on the other hand, avoids heat and better preserves thermolabile compounds, though it still demands considerable time and results in crude extracts that require further purification.



Emerging methods like enzyme-assisted extraction, supercritical fluid extraction, pressurized liquid extraction, ultrasound-assisted extraction, microwave-assisted extraction, and pulsed electric field extraction offer advantages in efficiency and selectivity but are cost-prohibitive for many research applications (Azmir et al., 2013).

Storage Effect

Storage is a critical factor in evaluating the effectiveness of bioactive compounds in fortified fermented dairy products. Over time, the efficacy of these compounds may diminish due to degradation or complex formation with milk proteins (Josipovid et al., 2015; Ozturkoglu-Budak et al., 2016). However, some studies have shown increased bioactivity post-storage, attributed to microbial breakdown and subsequent release of phenolics (Vicencsuto & de Castro, 2020a). Stabilization methods such as microencapsulation are being explored to preserve bioactivity during storage (Aramrueang et al., 2019).

Methodology of Investigation

The DPPH Radical Scavenging Assay is the most widely used method to assess antioxidant activity. In this assay, the purple DPPH radical is reduced to a colorless product by antioxidant compounds, typically measured at 515–528 nm. Additional techniques include the ABTS assay, Ferric Reducing Antioxidant Power (FRAP), and Ferric Ion Chelating (FIC) methods. Each method evaluates different aspects of antioxidant capacity, ranging from electron donation (DPPH, FRAP) to metal ion chelation (FIC), providing a comprehensive understanding of antioxidant potential

Conclusion

This review aimed to evaluate the effects of bioactive plant compounds on fermented dairy products. The findings highlight the significant potential of incorporating these compounds into products such as yogurt and cheese, which are widely consumed and therefore ideal vehicles for nutritional enhancement. Among the various sources, spices and fruits emerged as the most commonly used natural carriers of bioactive compounds, largely due to their favorable sensory properties. Furthermore, the antioxidant effects of these compounds have been extensively investigated, attributed to the high phenolic content present in many plant sources. Future research could focus on elucidating the specific interactions between bioactive compounds and fermented dairy matrices, which would help to better differentiate their functional and sensory effects.

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