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From tradition to innovation: dadih, the Minangkabau tribe's traditional fermented buffalo milk from Indonesia

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Abstract

Dadih, also known as *dadiah*, is a traditional fermented buffalo milk from Minangkabau tribe in West Sumatra. This product is similar to yogurt, with a smooth and shiny surface, thick-bodied, creamy color and pleasant flavor. Microbes that play a dominant role in dadih fermentation are lactic acid bacteria, which possess several functional features, including antimicrobial, antioxidant, antimutagenic, hypocholesterolemic and immunomodulatory properties. However, traditional dadih production faced numerous challenges that impede its development, including the limitation of buffalo milk production, unstandardized dadih production due to the spontaneous fermentation, safety concerns from the absence of heat treatment, as well as limited shelf-life and consumer acceptance of traditional dadih product. Therefore, it is crucial to improve buffalo cultivation, using different types of milk, and apply pasteurization process in dadih production. Furthermore, innovation in dadih product that is more durable and preferred by consumers can be achieved through the application of various processing and packaging technologies.

Keywords Buffalo milk, Dadih, Fermented food, Minangkabau, Yogurt

Introduction

Indonesia is the largest archipelago with abundant megabiodiversity, possessing a wealth of ethnic and cultural diversity. It is known as well for its unique cuisine and traditional fermented delicacies [1]. More than 1300 ethnic groups are represented in Indonesia, and each region or ethnic group has its own distinct traditional cuisine with its own history, purposes, ingredients and flavors [2]. As historical evidence or a symbol, traditional cuisine can be used to distinguish one ethnic group from another. Dadih is known as the authentic traditional food originating from Minangkabau (Minang), one of the ethnic groups in Indonesia, predominantly residing in the province of West Sumatra (Fig. 1) [2]. Apart from dadih, other authentic traditional foods such as *lamang* [3], *lamang tapai* [4] and *rendang* [5], also originated from this region.

Dadih is typical Minangkabau dairy product that is produced from buffalo milk through the application of natural fermentation methods [6]. Buffalo milk is fermented in bamboo tube with conditions that tend to be facultative anaerobic and use banana leaves as a cover [7]. It serves as a significant dairy product in the diet of the Minangkabau, resembling yogurt and is similar to *dahi* of India [8]. Dadih is relatively unpopular in comparison with other fermented milk products such as cheese, yogurt and kefir, due to the fact that it is manufactured

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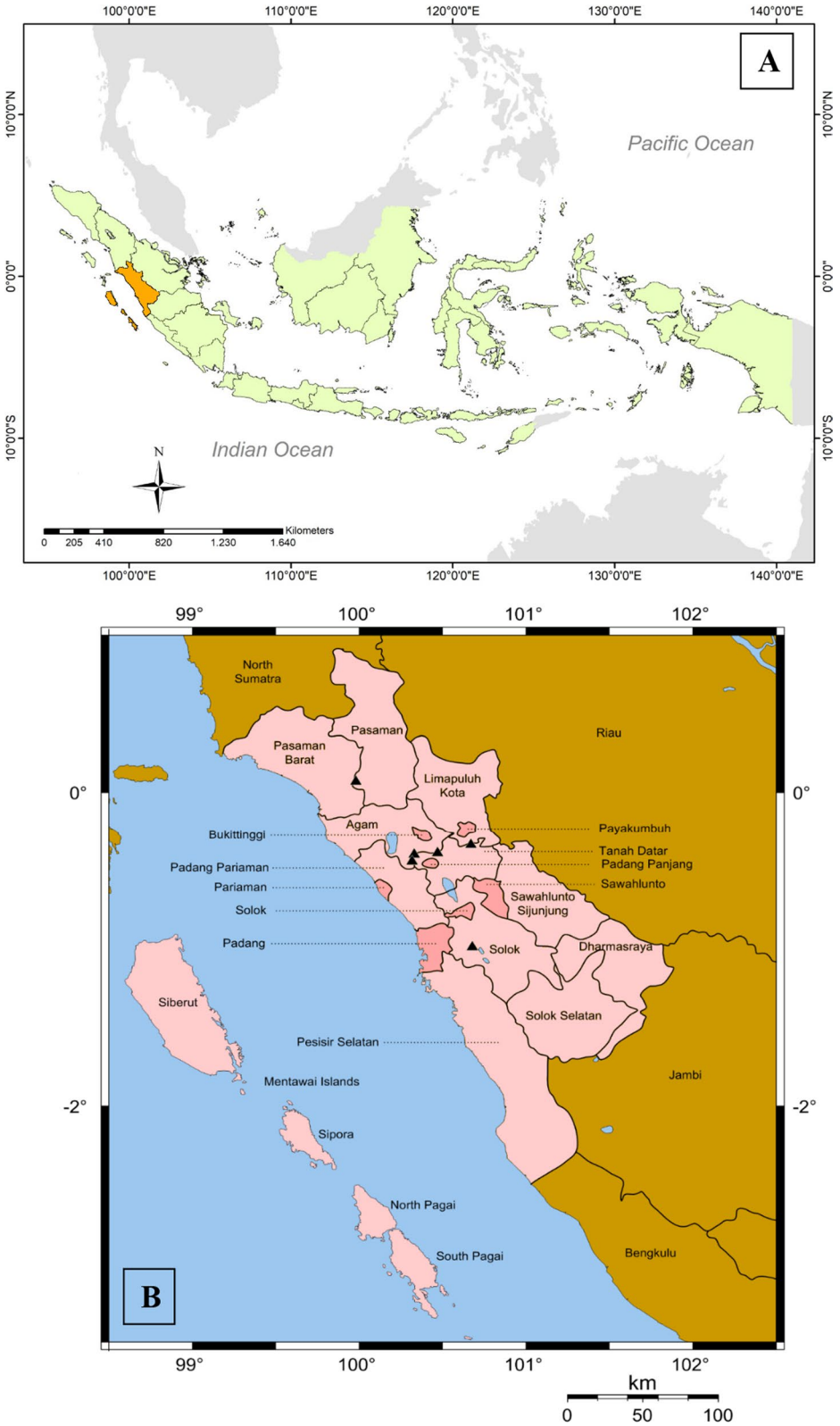

Fig. 1 **A** The geographical representation of West Sumatra Province in Indonesia, renowned for being the residence of the Minangkabau ethnic groups, **B** Dadih is popular dairy product in West Sumatra, especially in Tanah Datar, Lima Puluh Kota, Padang Panjang, Solok and Bukittinggi (retrieved from: https://images.app.goo.gl/dkBUQFzyumsPrdVx9)

traditionally [9]. Dadih production typically undergoes spontaneous fermentation, devoid of any starter culture inoculation, heat treatment or pasteurization [10]. The formation of dadih is facilitated by the presence of indigenous enzymes of buffalo milk, and naturally occurring lactic acid bacteria (LAB) of the buffalo milk, bamboo tubes and banana leaves involved in milk fermentation [11]. It has been observed that natural indigenous LAB derived from buffalo milk contributes the most, while bamboo tubes and banana leaves as well as personal hygiene practice may also contribute [1]. Because of this spontaneous fermentation, the indigenous LAB may vary from one place to place and over the time [9, 11]. Although excellent hygiene practices were not implemented during the production of dadih, the presence of the natural LAB in this product enhances its safety [11].

The health benefits of dadih have been demonstrated in a number of preclinical studies and anecdotal reports [12]. Multiple probiotic strains have been isolated from dadih, which might contribute to its health benefits [13]. Nevertheless, the precise mechanism by which dadih exerts its health benefits remains mainly unknown. Dadih is recognized as a potential probiotic due to its high concentration of LAB (10^8 cfu/g) [10, 14]. LAB has the ability to support the intestinal wall and provides numerous health advantages to the host [15]. The LAB derived from dadih has also been documented to possess various beneficial properties, including antimicrobial [15], hypocholesterolemic [16–18], antimutagenic [19], antioxidant [20], immunomodulatory [21, 22] and anti-stress [23, 24].

Numerous obstacles were nonetheless identified in dadih production, prompting the local government and numerous researchers to collaborate in an effort to identify the optimal resolution. However, the objective of this article is to elaborate various aspects of dadih, including historical context, production process and its microbiological, nutritional and functional properties. The safety aspects as well as future challenges and prospect of dadih are also discussed in this article.

Ethnic and history

Dadih is known as an indigenous traditional food that originated from one of the well-known ethnic groups in Indonesia, Minangkabau, West Sumatra [6]. Dadih, fermented buffalo milk in a bamboo tube, has been prepared and consumed by the Minang people for centuries [25]. It is a yoghurt-like product with a glossy and smooth surface, even consistency, creamy-white color, pleasant flavor and sour taste [1] (Fig. 2). The Minang people refer to it as *dadiah*, and it is readily available in the regions of Tanah Datar, Lima Puluh Kota, Padang Panjang, Solok and Bukittinggi (Fig. 1). Dadih has become an important part of Minangkabau food culture with its popularity beyond the borders of West Sumatra. Consequently, dadih is also present in provinces that border West Sumatra directly, including the Kampar region of Riau Province and the Kerinci region of Jambi Province [26]. These regions are believed to be greatly influenced by the presence of the Minangkabau ethnic group, who reside outside of West Sumatra [26].

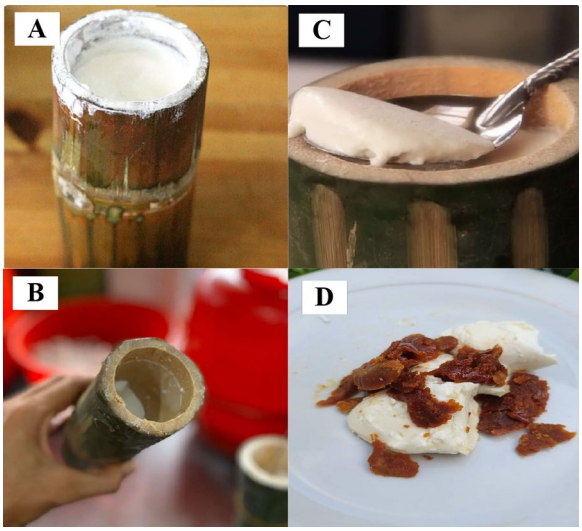

Fig. 2 **A**, **B** Dadih, also known as *dadiah, is* the authentic traditional fermented buffalo milk in bamboo container from Minangkabau, **C** Dadih has a creamy-white color, smooth and glossy surface, as well as thick consistency, **D** Dadih is generally served as a side dish with palm sugar as the topping

Dadih was historically discovered by chance when the Minangkabau tribe exposed bamboo containers containing milk to the sun. This process imparts a distinct acidic flavor to the milk and inadvertently thickens its consistency [27]. The most popular of consuming this cuisine is by pairing it with rice chips (*ampiang*), often referred to *ampiang dadih*. Dadih may also be consumed as a side dish with chili and onions. This combination produces a fresh sour-spicy taste as an accompaniment to rice [28]. Nowadays, dadih is usually served as a traditional delicacy at weddings and traditional ceremonies during the awarding of the honorary title of *Datuk* in West Sumatra [11].

Buffalo milk is the main raw material for dadih production. The milk used to produce traditional dadih comes from domestic buffalo or swamp buffalo (*Bubalus bubalis*). This type of buffalo has black body color and horns bent backward (Fig. 3). Their population in Indonesia has almost reached 90%, due to remarkable adaptability in acclimatizing to various environments [29]. For centuries, the buffalo has been a significant animal for the Minang people. The local people employed buffaloes for various uses, including transportation, delivery of goods,

Fig. 3 Domestic buffalo or swamp buffalo (*Bubalus bubalis*) is the type of buffalo with the largest population in Indonesia. This buffalo has black body color with horns bent backward, and their milk is usually fermented to produce dadih (retrieved from: https://images.app.goo.gl/UpwjAKrwJ2bL8you5)

ploughing fields, sugar cane grinding in the sugar production, and tourism activities such as buffalo fighting or *adu kerbau* [9]. Apart from their milk, the products from buffalo are also widely consumed, for example, as a source of meat and skin cracker production, while the excrement of buffalo is used for organic fertilizer and biogas production [9].

The limited production of buffalo milk is an obstacle to providing raw materials for making dadih. The milk produced is, of course, prioritized for newborn buffalo calves, and only part of the production can be used to make dadih. Buffalo rearing management that is not yet professional and is still far from modernization also causes low buffalo milk production in West Sumatra [26]. The amount of buffalo milk production in West Sumatra, which is only 2.40 L/head/day, is still challenging to develop the large amounts of dadih production [30]. Therefore, dadih is developed by using raw materials other than buffalo milk [26]. Dadih production can contribute to the Minang people's food security and the local economy [9]. Additionally, for the Minang people, dadih also represents the family host's emotional response toward their guests [31].

Dadih processing

The main materials used in dadih production are buffalo milk, bamboo tubes and banana leaves. Dadih is made by letting buffalo milk naturally ferment in a bamboo tube that is covered with banana leaves [1]. These leaves create conditions that are facultatively anaerobic, which is ideal for the fermentation process [23, 32, 33]. Alternately, plastic or taro leaves may also be utilized to cover the bamboo [34]. However, the use of different packaging materials of dadih affected its organoleptic properties, such as color, aroma, taste, and compactness, and texture properties [35]. Therefore, their development needs to be investigated further. For securing the cover, either banana bark or rubber bands are used [26]. Bamboo tubes are selected as fermentation containers with hygroscopic properties, which prevent whey separation in the product [1]. Bamboo's hygroscopic properties can prevent syneresis (water escaping the gel) by absorbing water, resulting in the thickening of buffalo milk into curd [36]. Various types of bamboo can be used in dadih fermentation [9, 37], such as bamboo ampel (*Bambusa vulgaris*), bamboo gombong (*Gigantochloa verticillata*), bamboo lengka tali (*Gigantochloa hasskarliana*), bamboo ater (*Gigantochloa atter*) and bamboo betung (*Dendrocalamus asper*) (Table 1). The native Minang people prefer to use bamboo ampel and bamboo gombong due to the bitter taste of these bamboo, so that the processed milk will not be contaminated by ants [38].

The production process of traditional dadih can be shown in Fig. 4. During the manufacturing process, the untreated buffalo milk (which is devoid of antibiotics) is filtered immediately to minimize the presence of physical impurities like stones and grass. Subsequently, the filtered milk is transferred into the bamboo tube, covered with banana leaves, and tied with a rubber band or banana bark [1]. Fermentation takes place naturally at ambient temperature (28–30 °C) for a duration of 24 h [11] or up to 2–3 days [16, 18]. The indigenous lactic acid bacteria (LAB) and proteases will carry out lactic acid fermentation and proteolytic activity during fermentation. Natural microorganisms in bamboo tubes also play an important role in breaking milk lactose down into organic acids [37]. As a result of the formation of organic acids, the pH of the medium will decrease, and the buffalo milk will then coagulate into a semi-solid consistency with a sour taste, known as dadih. The dadih that is produced will ascend to the surface, whereas the liquid will remain at the bottom [39–41]. LAB fermentation is an important part of making dadih because it lowers the amount of pathogenic contaminants and spoilage bacteria by making bacteriocins and organic acids, which stop the growth of these bacteria [42]. This makes dadih safer to consume.

In comparison with yogurt, heat treatment is not applied to the raw buffalo milk and no starter culture is added for dadih fermentation. The fermentation time of dadih is also longer (24 h up to 3 days) than yogurt (4 h). Damayanti et al. (2020) reported that the duration of fermentation influenced the characteristics of dadih. Increasing the duration of fermentation will lead to a denser consistency of dadih while also intensifying its sour taste. After conducting observations on three different fermentation times (24, 48 and 72 h), it was found that dadih fermented for 48 h is the most favored

Table 1 Types of bamboo in dadih production

Types of bamboo	Figure	Characteristics	References
Bamboo ampel (*Bambusa vulgaris*)		*B. vulgaris* is a 15–20-m-tall, erect, evergreen, clump-forming bamboo. It forms thorn-free, loose clumps, dark green leaves and lemon-yellow stems that are predominantly striped with green. The stems are initially tough, not straight or can split easily, stiff in nature, and have thick walls with narrow lanceolate leaves. The densely tufted stems can attain heights of 10–20 m and have a diameter of 4–10 cm	[93]
Bamboo gombong (*Gigantochloa verticillata*)		*Gigantochloa verticillata* is a type of large- to medium-sized bamboo with high water absorption capacity. It has erect stems with curved ends with yellow stripes when the bamboo is old. The leaves are green with an elongated lanceolate shape and a hairy leaf surface. This bamboo can grow up to 26 m with a bamboo stem circumference ranging from 25 to 30 cm	[94]
Bamboo ater (*Gigantochloa atter*)		*Gigantochloa atter* has light brown stem nodes, sometimes flanked by circular white lines on the stem nodes. The base of the ater bamboo stem is straight and the ater bamboo stem is bright green to dark green and can grow up to 12–15 m. Ater bamboo leaves are green and 30–35 cm long. The length of ater bamboo stem segments ranges from 30 to 40 cm with a bamboo stem circumference ranging from 25 to 30 cm	[94]
Bamboo lengka tali (*Gigantochloa hasskarliana*)		*Gigantochloa hasskarliana* is a type of bamboo with reeds reaching 10 m high, upright and straight. Young reeds are covered with black to brown hairs, when old they are slightly whitish green, the segments are 27–51 cm long, with a diameter of 3–6 cm, the walls are up to 10 mm thick. Green leaves with a length of 8–35 cm	[95]
Bamboo betung (*Dendrocalamus asper*)		*Dendrocalamus asper* is a type of bamboo that can grow up to 20–30 m. Betung bamboo stems are green, dark green or brownish green. Betung bamboo leaves have a smooth surface with white silk all over the leaf blade with a leaf length ranging from 35 to 42 cm. Betung bamboo stem segments are grayish green with a bamboo stem circumference ranging from 39 to 45 cm	[94]

by consumers due to its relatively smooth texture and mild sourness in taste [43].Furthermore, dadih fermentation involves mesophilic LAB at 28–30 °C, whereas yogurt fermentation involves thermophilic LAB at 45 °C [25]. Based on its consistency, the viscosity of dadih (349 cP) usually exceeds that of yogurt (324 cP), due to phase separation between dadih and some of the liquid part [44]. Dadih has a distinct aroma of buffalo milk that is absent in yoghurt. Dadih is more flavorful than yogurt due to its higher fat content, which is 6.48% compared to yogurt (3.25% fat). The higher the fat content enrich the flavor developed in the dadih products [1]. The involvement of indigenous yeasts in dadih fermentation also differs dadih from yogurt [45].

Microbiological properties

A wide range of microbiotas, including LAB and yeasts, may be present in products that ferment spontaneously. The range of viable LAB counts in dadih was between 1.42×10^8 and 3.80×10^8 cfu/g [46]. Microbes that play a dominant role in dadih fermentation are lactic acid bacteria such as *Streptococcus thermophilus, Lactobacillus bulgaricus, Lactobacillus casei, Bifidobacterium bifidum, Leuconostoc mesenteroides, Enterococcus liquefaciens, Lactococcus lactis*. These microbes can come from buffalo milk, bamboo tubes or banana leaves in the dadih fermentation process [47, 48]. Diverse varieties of LAB can be observed in different locations throughout West Sumatra [49]. Research on various traditional commercial dadih sold in Bukittinggi,

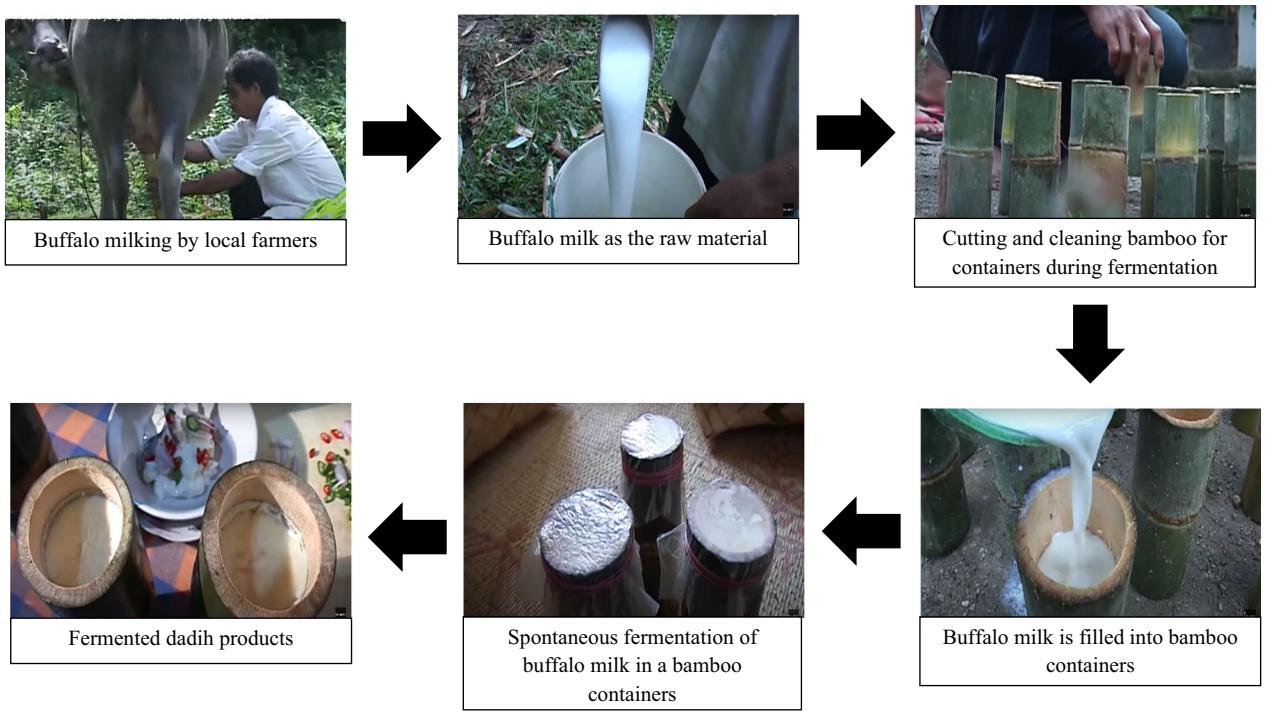

Fig. 4 The general production process of traditional dadih in West Sumatra, involving naturally fermented of buffalo milk in a bamboo container

Batusangkar, Lima Puluh Kota and Solok districts identified 10 species of LAB, including 4 species from the genus *Lactobacillus* sp. (*L. brevis, L. viridescens, L. buchneri* and *L. plantarum*), 2 species from the genus *Leuconostoc* (*L. mesenteroides* and *L. paramesenteroides*), 3 species from the genus *Streptococcus* (*S. lactis* subsp. *diacetylactis*, *S. raffinolactis* and *S. faecium*) and 1 species from the genus *Lactococcus* sp. (*L. piscium*) [49, 50]. These bacteria produce volatile chemicals, such as acetic acid, diacetyl and other aromatic molecules [51]. According to a study conducted by Nuraida [47], *Bacillus brevis*, *Leuconostoc mesenteroides*, *Lactococcus lactis* subsp. *lactis*, *Enterococcus faecium*, *Lactobacillus fermentum*, *Lacticaseibacillus rhamnosus* and *Lacticaseibacillus casei* were identified in dadih derived from buffalo milk. Dadih also contained *Enterococci* group bacteria, specifically *Enterococcus faecalis*, which could have been generated by an unsanitary process. This is likely the main contributing factor [32]. In Table 2, the numerous types of LAB found in dadih are described in detail.

In addition to LAB, various other varieties of microbes were identified in dadih. Dadih was found to have a greater abundance of species belonging to the orders *Burkholderiales* and *Serratia*, and a lesser abundance of *Brachybacterium* [48]. In dadih, three species of yeast were identified, *Candida metapsilosis*, *Kluyveromyces marxianus* and *Saccharomyces cerevisiae*. Among these species, *C. metapsilosis* was the most prevalent [52]. *Candida* sp., including *C. metapsilosis*, are commonly encountered in the gastrointestinal tract. However, they have also acquired the reputation of opportunistic pathogens and could potentially be the source of disseminated candidiasis [53]. *C. metapsilosis* was also isolated in the inner part of bamboo tubes in dadih [52]. Yurliasni [54], from the yeast identification, carried out obtained three species of yeast in dadih, namely *Candida curiosa, Brettanomyces custersii* and *Kluyueromyces lactis*. The presence of various yeast species, including *Candida stelimalicola* and *Pichia jadinii*, was also reported in dadih [55]. In a more recent study conducted by Farhana [56], various species of bacteria and yeast were found to be present in dadih. The bacteria mentioned are *Lactobacillus delbrueckii*, *Lactobacillus lactis* and *Lactobacillus acidophilus*. In addition to these species of lactic acid bacteria, yeasts have also been detected in dadih, including *Geotrichum* sp., *Saccharomyces* sp. and *Zygosaccharomyces* sp. [57]. The existence of yeasts in dadih fermentation should be considered, because it makes a positive contribution during fermentation process and the final products, such as providing growth factors for microorganisms and also producing flavoring substances [58]. Concerning

Table 2 Microorganisms found in dadih from different regions in West Sumatra

Region	Microorganisms	References
Lima Puluh Kota, Agam, Tanah Datar, Solok	*Lactobacillus brevis, Weissella viridescens, LentiLactobacillus buchneri, Lactiplanti bacillus plantarum* subsp. *plantarum, Leuconostoc mesenteroides, Leuconostoc paramesenteroides, Streptococcus lactis* subsp. *diacetylactis, Streptococcus faecium, Streptococcus raffinolactis, Lactococcus piscium*	[96]
Sijunjung	*Lactoplantibacillus plantarum* subsp. *plantarum*	[62]
Solok	*Lactoplantibacillus plantarum* subsp. *plantarum*	[97]
Bukittinggi	*Lactococcus lactis* subsp. *lactis, LeviLactobacillus brevis, Lactoplantibacillus plantarum* subsp. *plantarum, Lacticaseibacillus casei, Lacticaseibacillus paracasei, Leuconostoc mesenteroides*	[18]
Bukittinggi and Padang Panjang	*Lactobacillus* sp., *Lactococcus* sp., *and Leuconostoc* sp.	[46]
Kamang	*Lactobacillus pentosus, Lactococcus lactis* subsp. *lactis, Pediococcus pentosaceus, Lactococcus lactis* subsp. *cremoris, Lactoplantibacillus plantarum* subsp. *plantarum*	[50]
Payakumbuh	*Lactobacillus* sp., *Lactoplantibacillus plantarum* subsp. *plantarum*	[98]
Gadut	*Lactoplantibacillus plantarum* subsp. *plantarum, Lactococcus lactis* subsp. *cremoris, Lactococcus lactis* subsp. *lactis*	[50]
Palupuh	*Lactobacillus* sp., *Lactococcus* sp., *Leuconostoc* sp.	[48]

safety, some yeasts also produce natural killer factors that can inhibit the growth of undesired microorganisms. The interaction between yeasts and other microorganisms especially LAB brings advantages to the quality and safety of final products [59]. The diversity of types of microorganisms found in dadih is thought to be influenced by variations in the use of existing bamboo tubes [37, 41]. Differences in bamboo growing locations, bamboo species and ages are most likely factors in the variation in the levels and varieties of microorganisms dadih [41, 60]. Several investigations have also attempted to produce dadih by combining purified dadih cultures with evaporated buffalo or cow milk [61]. The purpose of this investigation is to assess the efficacy and physicochemical properties of dadih produced by various species of microorganisms, including those utilizing *Lactobacillus acidophilus* and *Lactobacillus casei* starters [40, 61].

Nutritional properties

Dadih is made from buffalo milk as the main raw material. Buffalo milk has a lower water content compared to cow's milk, but it has a greater total solid content, particularly in terms of fat and protein [40, 61]. Buffalo milk also contained 0.6% w/v whey protein and 3.2% w/v casein [1]. These characteristics create dadih produced from buffalo milk has semi-solid consistency after fermented in bamboo tubes by natural microorganisms [40]. Dadih has a higher dry matter concentration compared to buffalo milk. This is inextricably linked to the coagulation process that occurs during fermentation, which separates the majority of the water content in the milk [40, 61]. Buffalo milk typically has a higher concentration of total solids compared to milk from other mammals (Table 3). This is mostly due to the higher content of casein and fat, which give dadih its creamy taste, and thick-bodied consistency [16].

Previous research has reported the nutritional composition and chemical analysis of dadih from different

Table 3 Composition of mammalian milks for produce dadih

Type of mammals	Total solid (%w/v)	Ash (% w/v)	Fat (% w/v)	Whey protein (%w/v)	Casein (% w/v)	Lactose (% w/v)	References
Buffalo	17.2	0.80	7.40	0.60	3.20	4.80	[1]
Cow	12.7	0.70	3.70	0.60	2.80	4.80	[1]
Sow	18.8	ND	6.80	2.0	2.80	5.5	[1]
Sheep	19.3	1.0	7.40	0.90	4.60	4.80	[1]
Goat	13.2	0.80	4.50	0.40	2.50	4.10	[1]
Camel	15.0	0.70	5.40	1.00	2.90	5.10	[99]
Mare	15.0	0.70	6.80	2.00	2.80	6.20	[1]

ND not determined

Table 4 Nutritional properties of dadih from different regions in West Sumatra

Nutritional properties	Lintau	Sijujung	Tanah Datar		Solok	Agam		
Moisture (%)	73.71	75.5	76.7	66.1	81.8	61.9	82.4	73.6
Ash (%)	ND	0.68	ND	0.72	0.92	1.14	0.91	ND
Fat (%)	5.80	6.50	6.92	5.70	7.89	18.0	8.17	5.53
Protein (%)	8.14	5.01	6.31	12.4	6.91	10.9	7.06	7.96
Carbohydrate (%)	ND	12.3	ND	15.1	2.48	8.06	1.46	ND
pH	5.20	4.74	4.60	4.55	4.76	4.33	4.80	5.16
Acidity	ND	1.17	1.4	0.51	1.32	1.70	1.28	ND
Reference	[100]	[39]	[101]	[14]	[39]	[14]	[39]	[102]

ND not determined

regions in West Sumatra, which are detailed in Table 4. The nutritional composition and chemical analysis of dadih varied throughout different regions. For example, the type of buffalo feed may influence the pH and acidity of dadih. The buffaloes in Agam were fed with *Imperata cylindrica* grass, while in Tanah Datar, they were fed with banto grass (*Leersia hexandra* Sw.) mixed with rice straws. The presence of several compounds, including malic and citric acid, in these weeds can potentially influence the pH and acidity levels of the resulting dairy. Another study by Wirawati et al. [50] determined the characteristics of dadih in five districts of West Sumatra. The nutritional content of dadih was determined to be 4.08–4.52% protein, 8.47–9.32% fat, 17.82–20.43% solids and 1.21–1.61% acidity. Further, Wirawati et al. [49] also performed a commercial analysis of dadih, collecting samples from two specific locations in West Sumatra: Tilatang Kamang in Agam Regency and Lembah Gumanti in Solok Regency. Based on this research, the dadih composition consists of 81.79–82.40% moisture content, 6.91–7.06% protein, 7.98–8.17% fat, 0.90–0.91% ash and a pH range of 4.76–4.81. These studies show a normal level of protein, usually more than 4%. The variations in the characteristics of dadih are believed to be influenced by significant factors pertaining to the raw materials and manufacturing process. These factors include the initial composition of buffalo milk, the type (e.g., thickness, porosity) and size of the bamboo tube, as well as the duration of the buffalo milk fermentation process [41, 60]. The lack of regulation in the production process, resulting in varying chemical properties of dadih across different regions, has prompted the need for uniformity in its manufacture.

Several studies have documented the replacement of buffalo milk with alternative forms of milk as the substrate for dadih production (Table 5). In this scenario, the utilization of back-slopping method or the use of starter culture may be necessary to initiate the fermentation process and ensure standardization in the production process. The results showed that the fat content of dadih from buffalo milk (5.53–18.0%) (Table 4), was comparable to that of dadih from cow milk (7.03–11.0%) (Table 5), although the fat content of fresh cow milk is lower (Table 3). Some additional processes, such as evaporation and the addition of skim milk, can enhance the overall total solid content of cow milk, which includes fat and

Table 5 Nutritional properties of dadih from different types of milk

Nutritional properties	Soy milk	Goat milk	Cow milk		
Starter culture	Back-slopping method using dadih from Bukittinggi (4% w/w)	Spontaneous	Back-slopping method using dadih from Bukittinggi (4% w/w)	*L. plantarum* + *L. acidophilus* (3% w/v)	*L. plantarum* + *L. acidophilus* + *B. bifidum* (3% w/v)
Moisture (%)	ND	ND	ND	73.7	83.2
Ash (%)	ND	ND	ND	0.52	0.90
Fat (%)	2.91	ND	7.03	7.59	11.0
Protein (%)	4.65	3.75	9.79	4.27	3.53
Carbohydrate (%)	ND	ND	ND	13.9	1.37
pH	ND	6.54	ND	4.29	3.49
Acidity	ND	0.31	ND	0.64	3.45
Reference	[40]	[41]	[40]	[103]	[104]

ND not determined

protein, before it is used as substrate for dadih production to achieve the desired qualities of final dadih product [39]. However, the fat content of dadih from soy milk is still lower than that from other kinds of milk, despite that skim milk was added to increase the total solid content.

According to the study conducted by Hosono et al. [32], dadih made from buffalo milk using a 3% *Streptococcus lactis* starter contains a minimum of 16 different types of amino acids. Out of the amino acids that currently exist, there are ten that are considered essential such as threonine, valine, methionine, isoleucine, leucine, phenylalanine, tryptophan, histidine, lysine and arginine. Furthermore, Harnentis et al. [23] found dadih made from cow milks contains a total of 17 types amino acids, including proline and cystine, which were absent in dadih mae from buffalo milk. Regarding digestibility, Harnentis et al. [23] elaborated that the presence of microorganisms in dadih leads to a fermentation process that breaks down proteins into smaller components. The digestion potential of dadih protein is enhanced by the presence of human pepsin and pancreatin enzymes, leading to an improvement in its digestibility [40]. From studies conducted utilizing *S. lactis* in the creation of dadih from cow milk, the digestibility of dadih protein through laboratory testing (in vitro) was in the range of 95.22–98.56%, where there was a trend to enhance protein digestibility with an increasing percentage of starter added [40, 61].

Potential functional properties of dadih for health

Dadih demonstrated numerous potential health benefits due to its abundance of viable LAB, including antimicrobial, antioxidant, antimutagenic, immunomodulator, anticholesterol and a source of ***γ***-aminobutyric acid (GABA) (Table 6). Several factors contributed to the antimicrobial activity of LAB from dadih, including decreased pH levels, competition for substrates and the production of substances with bactericidal or bacteriostatic properties [62]. The probiotic *Lactobacillus* spp. found in dadih exhibited antimicrobial activity against *Escherichia coli*, *Staphylococcus aureus* and *Salmonella enteritidis*, with inhibitory diameters of 11.54 mm, 10.27 mm and 16.31 mm, respectively [63]. The *L. plantarum* isolated from dadih can survive in 0.5% bile salts and acidic media up to pH 2. This makes it a good probiotic that can inhibit the growth of pathogenic bacteria in the digestive system [64]. *L. plantarum* also produced plantaricin that showed antimicrobial properties against *Staphylococcus* spp. [65].

An investigation was conducted into the antimicrobial activities of *L. plantarum* in dadih from the Solok, Sijunjung and Payakumbuh regions against *S. aureus, E. coli*, and *S. typhi*. The results showed that *L. plantarum* in dadih from Payakumbuh exhibited the strongest antimicrobial activity [66]. The mortality percentages of 33.3% for *S. aureus*, 27.27% for *E. faecalis* and 23.07% for *E. coli* were reported after 7 h of contact with *L. plantarum* in dadih from the Payakumbuh region. These results indicate that *L. plantarum* inhibits Gram-positive bacteria (*S. aureus* and *E. faecalis*) more effectively than Gram-negative bacteria (*E. coli*) [64]. This could be attributed to the outer membrane of Gram-negative bacteria, which acts as a highly effective barrier to permeability and can prevent the entry of large molecules (such as bacteriocins or enzymes) and hydrophobic compounds (such as hydrophobic antibiotics) [67]. Collado et al. (2007) also analyzed the adhesion capacity of specific LAB isolates from dadih to intestinal mucus and assessed their ability to inhibit model human pathogens. The results showed that all tested LAB strains displaced and inhibited pathogen adhesion, with *L. plantarum* IS-10506 showing the most adhesive and reduced pathogen adhesion to mucus [68].

Kusumaningtyas and Utami (2020) determine the antioxidant activities of dadih from different regencies in West Sumatra by using ABTS, DPPH and Fe-reducing power methods. The result showed that the soluble protein of dadih from the Agam region had the highest antioxidant activities in terms of Fe-reducing power, and scavenging activity against ABTS and DPPH [69]. In vivo studies using experimental rats showed that the groups given dadih once a day and twice a day had antioxidant properties by reducing the levels of malondialdehyde (MDA) in kidney tissue and reducing renal interstitial fibrosis rank in aging kidneys compared to the control group. The group given dadih twice a day showed the highest reduction in average fibrosis rank [70]. Histological observation of the mice's pancreatic organs was also carried out to determine the antidiabetic effect of dadih. The results showed that giving dadih can improve the pancreas of mice with diabetes mellitus, as seen from the histopathological picture of those who experiences pycnosis [71].

Pato et al. (2019) reported the effect of *Lactobacillus casei* subsp. *casei* R-68 (LCR68) isolated from dadih on the procarcinogenic enzyme activity and fecal microflora count of rats challenged with pathogenic bacteria. The results showed significantly higher counts of lactic acid bacteria and lower counts of *Escherichia coli* after the application of fermented milk LCR68. The presence of LCR68 in fermented milk reduced the activity of β-glucuronidase and β-glucosidase significantly in the feces of Wistar rats, so it is expected to be able to prevent the formation of procarcinogenic compounds into carcinogens [72]. The LAB isolates from dadih such as *L. paramesenteroides* R-62 and R-8, *Streptococcus lactis* subsp. *diacetylactis* R-63 and *Streptococcus cremoris*

Table 6 The potential functional properties of dadih

Functional properties	Method of analysis	Result	References
Antimicrobial	In vitro	The probiotic *Lactobacillus* spp. isolated from dadih exhibited potent antimicrobial effects against *Escherichia coli, Staphylococcus aureus* and *Salmonella enteritidis*	[63]
	In vitro	The probiotic *L. plantarum* strain 8m-21 isolated from dadih in the Solok region exhibited antimicrobial activity against *Escherichia coli* O157. Their inhibitory zone was greater than penicillin, ampicillin and kanamycin	[15]
	In vitro	The probiotic *L. plantarum* strain BDL11 isolated from soymilk dadih exhibited highest bacteriocin activity against *Listeria monocytogenes* compared to other strains	[87]
	In vitro	The probiotic *L. plantarum* isolated from dadih exhibited resistance to acidic media (until pH of 2) and bile salts (0.5%), so it can inhibit the growth of pathogenic bacteria in the digestive tract	[64]
	In vitro	The probiotic *L. plantarum* isolated from dadih in the Payakumbuh region exhibited greater antimicrobial properties than dadih from the Solok and Sijunjung regions against *Escherichia coli*, *Staphylococcus aureus* and *Salmonella typhi*	[66]
	In vitro	Plantaricin produced by *L. plantarum*, which was isolated from dadih in the Payakumbuh region, inhibited the growth of Gram-positive bacteria (*S. aureus* and *E. faecalis*) more effectively than Gram-negative bacteria (*E. coli*)	[39, 64]
	In vivo	*Enterococcus faecium* (strains IS-16183, IS-23427 and IS-27526), and *L. plantarum* (strains IS-20506 and IS-10506) isolated from dadih were able to significantly reduce the adhesion levels of pathogens (*Bacteroides vulgatus, Clostridium histolyticum, Escherichia coli, Salmonella* Typhimurium and *Staphylococcus aureus*). All pathogens were also displaced by natural strains from dadih. *L. plantarum* strain IS-10506 showed the most adhesive to human intestinal mucus and reduced human pathogen adhesion	[68]
Antioxidant	In vitro	Dadih produced by fermenting cow milk in a 1:1 ratio with 1% starter cultures of *L. casei* and *L. plantarum*, exhibited antioxidant activity by scavenging DPPH radicals	[20]
	In vitro	The soluble protein of dadih (fermented for 3 days) from the Agam region exhibited highest antioxidant properties against DPPH and ABTS radicals, as well as showed Fe-reducing power	[69]
	In vivo	Giving dadih to experimental rats showed antioxidant properties by reducing the levels of malondialdehyde (MDA) in kidney tissue and reduce renal interstitial fibrosis rank in aging kidneys	[70]
Antidiabetic	In vivo	Giving dadih can improve the pancreas of mice with diabetes mellitus based on the histopathological picture of those who experiences pycnosis	[71]
Antimutagenic	In vivo	Fermented milk added with *Lactobacillus casei* subsp. *casei* R-68 (LCR68) isolated from dadih can significantly reduce the activity of β-glucuronidase and β-glucosidase in Wistar rat feces, and expected to prevent the formation of procarcinogenic compounds into carcinogens	[72]
	In vitro	Several LAB strains isolated from dadih, including *Leuconostoc paramesenteroides* R-62 and R-8, *Streptococcus lactis* subsp. *diacetylactis* R-63 and *Streptococcus cremoris* R-48, were found to exhibit strong antimutagenic properties against various mutagens such as N-nitroso-dimethylamine (NDMA), N-nitroso-diethylamine (NDEA), N-nitroso-piperidine (NPIP) and N-nitrosopyrrolidine (NPYR)	[73]
	In vivo	The milk cultured with *E. faecium* strain IS-27526 isolated from dadih in Bukittinggi significantly lowered the fecal mutagenicity of rats toward amino acid pyrolyzate (Trp-P1) mutagens	[19]
	In vitro	The milk cultured with *Lactococcus lactis* subsp. *cremoris* R-48, *Leuconostoc mesentroides* R-51 and *Lactococcus lactis* subsp. *casei* R-68 isolated from dadih, showed strong inhibition against the mutagenicity of both heated salty and sweet tauco	[105]
Immunomodulator	In vivo	The salivary secretory immunoglobulin A (sIgA) levels in underweight Indonesian preschool children significantly increased after 90 days of milk supplementation cultured with *E. faecium* IS-27626 isolated from dadih, at a dosage of 2.31×10^8 cfu/day	[21]
	In vivo	Supplementation of probiotic *L. plantarum* strain IS-10506 and zinc for 90 days in young children resulted significant increase of humoral immune response and improved their zinc status	[106]
	In vivo	Supplementation of *L. plantarum* strain IS-10506 isolated from dadih stimulates TGF-1 and causes an increase in sIgA (the first line of defense in protecting the intestinal epithelium from enteric toxins and pathogenic microorganisms) in children under two years old	[107]
Anticholesterol	In vitro	The LAB present in dadih can reduce serum cholesterol levels through direct binding to dietary cholesterol and/or deconjugation of bile salts	[75]
	In vivo	The rats fed with fermented milk *Lactococcus lactis* subsp. *lactis* IS-10285 isolated from dadih exhibited significantly lower total bile acids in their serum and may contribute to the reduction of cholesterol	[17]
GABA-source properties	In vitro	The LAB isolated from dadih, such as *Pediococcus* and *L. plantarum* N5, have been identified as potential producers of GABA (γ-aminobutyric acid)	[23, 24]

R-48 also have anticancer potential because they have antimutagenic properties through binding mechanisms of mutagenic compounds, such as N-nitrosodimethylamine (NDMA), N-nitrosodietilamin (NDEA), N-nitrosopiperidine (NPIP) and N-nitrosopyrrolidine (NPYR) [73]. In vivo antimutagenic properties of *E. faecium* IS-27526, isolated from dadih in Bukittinggi, toward amino acid pyrolyzate (Trp-P1) mutagens were also observed. Milk cultured with *Enterococcus faecium* IS-27526 significantly lowered the fecal mutagenicity of rats as compared to the control group [19]. The mechanism of the antimutagenic effect of LAB in dadih takes place because of the bonds between mutagens or carcinogens and the peptidoglycan contained in the LAB. Mutagens and carcinogens, which are bound, will be excreted via feces and urine [15].

A study documented the immunomodulatory properties of LAB in dadih. The levels of salivary secretory immunoglobulin A (sIgA) in underweight Indonesian preschool children were significantly increased ($p < 0.05$) after 90 days of supplementation with *E. faecium* IS-27626 from dadih origin. The supplementation involved a daily dose of 2.31×10^8 cfu in 125 mL of ultra-high temperature (UHT) low-fat milk [21]. Furthermore, there was also an apparent increase in weight among children who had a normal body weight, suggesting that *E. faecium* IS-27626 can enhance the integrity of the colon, hence facilitating optimal absorption of nutrients. Possible protective strategies against infections include adhesion to and colonization of mucosal surfaces, which involve competing for binding sites and nutrients, as well as immunological regulation [21]. The sIgA serves as the main defense mechanism in protecting the gut epithelium from enteric toxins and pathogenic microorganisms. The sIgA enhances the removal of antigens and pathogenic microorganisms from the intestinal lumen by preventing their interaction with epithelial receptors, trapping them in mucus and facilitating in their elimination through peristaltic and mucociliary processes. This process is known as immunological exclusion [74].

Previous research has been reported the cholesterol-lowering effects of dadih. The LAB strains isolated from dadih, *L. fermentum* I-11 and *L. lactis* subsp. *lactis* I-2775, have been recognized as promising probiotics for the prevention of coronary heart disease. This is attributed to their ability to tolerate acidic conditions and oxgall (bile), as well as their capacity to deconjugate sodium taurocholate and bind cholesterol [75]. In addition, *Lactococcus lactis* subsp. *lactis* IS-10285 and IS-29862 have shown their capacity to deconjugate taurocholate [18]. In rat studies, the rats that were given fermented milk cultured with *Lactococcus lactis* subsp. *lactis* IS-10285 from dadih showed a notable decrease in the levels of total bile acids in their blood serum [17]. The LAB present in dadih can modify serum cholesterol levels by either directly binding to dietary cholesterol, and/or by deconjugating bile salts [17]. The action of bile salt hydrolase may decrease cholesterol levels by making the free bile salts (deconjugated) less capable of being reabsorbed in the gut compared to the conjugated bile salts. As a result, these deconjugated bile salts may be expelled in the feces [18]. The LAB obtained from dadih have been identified as a potential producer of GABA (γ-aminobutyric acid), including *Pediococcus acidilactici* [24] and *L. plantarum* N5 [23]. GABA functions as the main inhibitory neurotransmitter in the central nervous system of mammals [76]. GABA has also been found to play a role in protecting broiler chicks from heat-induced stress [77]. Additional research is required to specifically determine the anti-stress effects of GABA-producing LAB found in dadih, particularly in human subjects.

Safety aspect and improving shelf life of dadih

The production process of dadih still involves spontaneous fermentation and conducted traditionally, so there is no control over procedures, substrates, storage, packaging and sanitation, which leads to the proliferation of spoilage or pathogenic microorganisms [78]. The hygienic quality of the substrates, ingredients, water, utensils, environmental conditions (e.g., temperature and humidity) and good hygiene practices may affect the microbial communities associated with the fermentation [79, 80]. A well-conducted fermentation generally guarantees the production of safe product for the consumer. The presence of natural LAB in dadih can help avoid foodborne diseases from poor hygienic conditions. The LAB initiates acidification in the raw materials (buffalo milk) by producing organic acids, aromatic compounds and bacteriocins to inhibit spoilage and pathogenic microorganisms [1], thus making dadih safer to consumer.

Traditional dadih product generally has a short shelf life, only 3 days at room temperature [81]. To extend its shelf life, dadih is usually stored at low temperatures (< 10 ℃) [39]. Dadih stored at low temperatures can have a shelf life of up to 30 days [82], indicating that low temperature is the optimal environment for storing dadih. However, low-temperature storage is highly conducive to the proliferation of psychrotrophic bacteria which can cause spoiling, such as *Alcaligenes* spp., *Pseudomonas* spp., *Achromobacter* spp., *Escherichia* spp. and *Micrococci* spp. These bacteria are aerobic and Gram-negative [83]. They have the ability to proliferate quickly at low temperatures around 7 °C and generate powerful proteolytic enzymes that degrade caseins, leading the curd to either liquefy or develop a slimy texture on the surface [83]. Other microorganisms that contribute to the deterioration of dadih

are yeasts, specifically *Rhodotorula* spp. and *Torulopsis* spp., as well as molds like *Geotrichum* spp., *Mucor* spp. and *Penicillium* spp. They are associated with surface defects of dadih, such as discoloration and liquefaction of the curd, or the formation of pigmented colonies [83]. Indeed, all of these microorganisms have a limited ability to withstand high temperatures and are effectively eliminated through pasteurization [83]. Therefore, the microbial burden of these spoilage microorganisms will be diminished through pasteurization of the raw material.

The quality of dadih is influenced by the duration of storage and temperature during storage. The longer the storage time will decrease the quality of dadih including taste, aroma, color, and preference [82]. The increasing temperature during storage will decrease the pH level at which spoilage microorganisms initiate their growth. At a temperature of 7 °C, the predominant growth of *Pseudomonas* spp. occurred at a pH of 4.80, whereas no growth was observed at a pH of 4.50. However, at a temperature of 20 °C, certain strains of *Pseudomonas* spp. were still able to grow at a pH of 4.40. Similarly, an increase in the storage temperature from 7 to 20 °C resulted in a reduction in pH at which *Enterobacter agglomerans* began to grow, from 3.80 to 3.60 [84]. From the above discussion, it can be concluded that hygienic production practices and application of low-temperature storage are suitable preservation methods that may be required to extend the shelf life of dadih.

Challenges, prospects and product innovation

Dadih production undergoes spontaneous fermentation, devoid of any starter culture inoculation, heat treatment or pasteurization [85]. Since dadih production does not involve any heat treatment, it fails to meet the requirements that mandate pasteurization of raw milk [1]. The curdling of fresh buffalo milk into dadih is a result of the existence of native enzymes and natural LAB in buffalo milk and the production environment [1]. As the sensory properties of dadih may differ due to various LAB in fermentation and due to spontaneous fermentation, standardizing the protocol of dadih production and microbiota contributed in dadih is necessary to increase the safety of dadih for human consumption. Pasteurization and controlled fermentation are also important to make dadih more significant in the food industry. Thickening, gelation and coagulation of buffalo milk during spontaneous fermentation of heated buffalo milk attributed to the proteolytic activity from either protease of bacterial origin or milk protease such as plasmin, which is heat-stable proteases [86].

Inadequate production of buffalo milk further complicates the procurement for dadih production. This constraint arises due to the declining buffalo population and the utilization of swamp buffalo as a non-dairy buffalo breed [26]. The use of dairy buffalo breed species for long-term use as a source of fresh milk is an additional endeavor that can be pursued [49, 66]. It could manifest upon application. The existence of dairy buffalo in Indonesia is confirmed by the occurrence of the Murrah buffalo, a type of river buffalo, in North Sumatra. This breed of buffalo exhibits higher milk production capacity and a longer lactation period compared to swamp buffalo [66]. This is a crucial aspect of their development in order to support dadih production. Considering the similar geographical and condition, the prospects for its growth in West Sumatra are highly favorable, especially in efforts to promote the cultivation of buffalo, and subsequent buffalo milk production and dadih manufacturing. Further consideration must also be given to undertakings such as cross-breeding, artificial insemination or the introduction of dairy buffalo into the buffalo breeder community, while also considering the regional conditions, challenges and future prospects [49, 66]. The limited quantity of buffalo milk in dadih production can be also substituted with other potential alternative ingredients which are more available, such as cow milk [49], goat milk [60] or even soymilk [87]. However, some additional processes, such as evaporation and skim milk addition, are needed to equal the total solid content of buffalo milk to produce dadih with desired properties [60, 88].

An additional area for potential improvement of dadih product may come from the packaging. Replacing dadih containers from bamboo to alternative containers with more sterile and hygienic (plastic or glass containers) is also important in commercialization and large-scale production [60]. In the study conducted by Sisriyenni and Zuriyati (2004), the shelf life of dadih made from buffalo milk was improved by substituting bamboo tubes to plastic tubes. The sensory quality of dadih from buffalo milk in plastic tubes is relatively not significantly different compared to dadih in bamboo tubes, both in terms of taste, aroma, color, consistency and consumer preferences. [89]. However, the development of plastic packaging needs to be investigated, as plastic waste may cause environmental problems. To support the marketing of dadih, developing packaging designs with complete labeling can also be applied. In order to increase the shelf life of dadih, innovative developments can be conducted by producing dadih powder using spray drying technology [90]. Spray drying is a significant process in the manufacturing of commercial powdered milk and can also be used to produce dadih in powder form. This method has the potential to develop a more standardized and controlled dadih product.

Current research on microbial communities in dadih still relies on the culture-based approaches. These

approaches have produced a limited amount of information because of the presence of many unculturable microorganisms and their ineffectiveness for diverse microbial communities. Culture-based techniques also require a large amount of time and material cost [91]. As such, culture-independent approaches, including high throughput sequencing (HTS) and next-generation sequencing, which can quantify hundreds of thousands of species inhabiting a microbial ecosystem [92], can be applied in the future research to provide an opportunity to uncover succession and dynamic of microbial community in dadih fermentation ecosystems.

Conclusion

Recent advancements led to an investigation of the attributes of traditional foods. Dadih, a traditional fermented buffalo milk, has been an integral part of Indonesian food culture, especially in the Minangkabau tribe. It contains a variety of natural microorganisms, particularly LAB microflora with probiotic properties, which contribute to its nutritional value and potential health advantages. Dadih fermentation is closely related to the local culture, which still utilizes spontaneous fermentation. Consequently, a wide variety of microbial populations were present in the product, which impaired the quality of the product. Standard protocols, fermentation process specifications and quality control for dadih must be formulated so that these products are safer for human consumption. The improvement of buffalo cultivation, use of alternative raw materials and product innovation can also be considered for product sustainability and commercialization. This review would be beneficial and encouraging for additional research regarding dadih.

Abbreviations

ABTS	2,20-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)
cfu	Colony forming unit
DPPH	2,2-Diphenyl-1-picryl-hydrazyl-hydrate
GABA	γ-Aminobutyric acid
LAB	Lactic acid bacteria
sIgA	Secretory immunoglobulin A

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Availability of data and materials

All data and materials are presented in the manuscript.

Declarations

Ethics approval and consent to participate

All authors declared that all photos in images 2 and 4 are personal documentation and property rights material from Dr. R. Haryo Bimo Setiarto, the corresponding author of this manuscript.

Consent for publication

All authors declared that all photos in images 2 and 4 are personal documentation and property rights material from Dr. R. Haryo Bimo Setiarto, the corresponding author of this manuscript.

Competing interest

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