



## REVIEW



# Impact of Milk Fat on Cheese Flavor and Novel Technologies for Its Enhancement

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Received: December 3, 2024  
Revised: December 12, 2024  
Accepted: December 12, 2024

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## Abstract

Milk fat plays a crucial role in the sensory properties of cheese, contributing to the development of a wide range of flavor compounds. During cheese ripening, various microbiological and biochemical processes drive the formation of these flavors. The microbial metabolism of milk fat is particularly important, as short- and medium-chain fatty acids significantly influence the characteristic flavor of cheese. Additionally, the use of exogenous lipases in cheese production can enhance flavor development and accelerate the ripening process. This review aims to examine the mechanisms by which milk fat contributes to flavor development during cheese ripening and to explore novel approaches for enhancing cheese flavor through microbiological and enzymatic pathways.

## Keywords

milk fat, cheese flavor, lipase, adjunct culture

## Introduction

The consumption of fluid milk per person in Korea has been decreased during the last 5 years due to a decrease in the birth rate and an increase in alternative beverage market [1]. However, the consumption of cheese has increased from 3.0 to 3.7 kg per person since cheese is an excellent source of protein and calcium providing additional health benefits to consumers [2,3]. In Korea, fresh cheese is mainly consumed rather than ripened cheese because some consumers do not prefer ripened cheese due to its unfamiliar and unpleasant flavor [4]. Flavor has been known as a critical sensory attribute to affect consumer's choice and preference [5,6]. Among the milk components, fat plays an important role in the sensory attributes of cheeses since it can contribute to the formation of various flavor compounds for cheese [7]. In order to further increase the consumer preference for cheese flavor and market size of cheese, it is critical to investigate how cheese flavors are developed. Recently, consumer demand for low fat and reduced fat cheeses has remained strong because of increasing consumer interest in health. However, a reduction in the fat content of cheese negatively affects cheese flavor. Therefore, various novel technologies enhancing the flavor of reduced and low-fat cheeses need to be reviewed.

## 1. Physicochemical and flavor properties of milk fat

### 1) Physicochemical properties of milk fat

Milk fats are present as milk fat globules, mostly 3-10  $\mu\text{m}$  in diameter, stabilized by milk fat globule membranes and are dispersed in the milk serum [8-10]. In bovine milk, the fat content has been known to be 3.5%-5% depending on the season, feed, lactation stage, and species [9,11,12]. For example, milk fat content tends to be higher in winter and lower in summer, which can be associated with feed intake [13]. In addition, when the grain intake of dairy cattle increased, the production of milk fat in mammary gland tissue decreased resulting in a lower milk fat content [14]. According to Palmquist, milk fat content decreased until the initial 40 to 60 days after calving [15]. It then increased until the late lactation, which was related to the yield of milk. The milk fat content was also varied depending on cattle breeds. For example, Jersey milk has a relatively high fat content compared to other varieties, such as Holstein and Guernsey [16,17].

### 2) The formation and characteristics of milk fat-derived flavor compounds in cheese

Milk fat is composed primarily of about 98% triglycerides consisting of glycerol and fatty acid [10]. Milk triglycerides are hydrolyzed by microbial and endogenous lipase to form fatty acids including short-chain, medium-chain, and free fatty acids [18]. Lipases are derived from raw milk, starter culture, secondary or non-starter culture, and rennet. Milk triglycerides are decomposed into fatty acids with short and intermediate chains and free fatty acids through hydrolysis by indigenous, endogenous and exogenous lipases during cheese ripening [18-20]. Short- and medium-chain fatty acids (i.e., 4-12 carbon atoms) contain lower flavor thresholds while long-chain fatty acids have higher flavor thresholds. It indicates that short- and medium-chain fatty acids have a greater role in the development of cheese flavors due to their lower thresholds compared with long-chain fatty acids [21]. During cheese ripening, the production of fatty acids with short- and medium-chains and an even number of carbon atoms (i.e., carbon number between 4 and 12) contributes to the characteristic cheese flavors. In the case of Cheddar cheese, butanoic acid as a principal volatile fatty acid was reported to contribute to the rancid cheese flavor, while hexanoic acid imparted cheesy, sweaty, fatty, and sour flavor [22,23]. According to Sablé et al., the butanoic acid plays an important role in the unique flavor characteristics of Camembert cheese, contributing to the rancid, cheesy, and sweaty flavor [24]. Octanoic acid is integral to the flavor attributes of Camembert cheese, imparting the goaty and fruity flavor while pentanoic acid contributes to cheesy like and sweaty flavor [24,25]. The blue-veined cheeses including blue cheese present goaty, pungent, and rancid flavors, which are attributed to the formation of butanoic, hexanoic, and octanoic acid [24]. The milk fat-derived flavor compounds in four different types of cheese (Gouda, Cheddar, Camembert, and Blue cheese) are presented in Table 1.

**Table 1.** Flavor compounds derived from milk fat in four types of cheese

	Compound	Classification	Flavor attribute	Reference
Gouda	Butyric acid	Fatty acid	Cheesy, rancid	[20], [26], [27]
	Butanon	Methyl ketone	Etheric, acetone	[20], [26]
	Heptanal	Aldehyde	Green, hay, stale	[20], [28]
	Octanal	Aldehyde	Green, hay, stale	[28]
	Pentanal	Aldehyde	Fragrant, sweet, fruity, green, leaf	[20], [27]
	$\delta$ -Decalactone	Lactone	Delicate, sweet, peach, coconut-like	[26], [28], [29]
	Ethyl butyrate	Ester	Fruity	[28]
Cheddar	Butyric acid	Fatty acid	Cheesy, rancid	[20], [26], [27]
	Acetic acid	Fatty acid	Sour	[20], [30]
	Hexanoic acid	Fatty acid	Sweet, acidic, cheesy, sharp, goaty	[30], [31]
	Decanoic acid	Fatty acid	Soapy, waxy	[31]
	1-Octen-3-one	Methyl ketone	Mushroom, metal	[20], [29], [30], [32], [33]
	2-Butanone	Methyl ketone	Etheric, acetone, sweet	[26], [27]
	$\delta$ -Dodecalactone	Lactone	Sweet, coconut-like	[30]
	$\gamma$ -Octanoic lactone	Lactone	Sweet, coconut-like	[30]
	$\delta$ -Octalactone	Lactone	Sweet, coconut-like	[30]
$\gamma$ -Dodecalactone	Lactone	Sweet, coconut-like	[30]	
Camembert	Butyric acid	Fatty acid	Cheesy, rancid	[20], [26], [27]
	1-Octen-3-ol	Methyl ketone	Mushroom, musty	[20], [29], [30], [32]
	1-Octen-3-one	Methyl ketone	Mushroom, metal	[33]
	$\gamma$ -Decalactone	Lactone	Peach, apricot, coconut-like	[5], [20], [26]
	$\delta$ -Decalactone	Lactone	Delicate, sweet, peach, coconut-like	[26], [28], [29]
	2-Heptanone	Methyl ketone	Mouldy flavor, spicy, cinamon	[34]
	2-Nonanone	Methyl ketone	Mouldy flavor, fruity, floral	[34]
Blue-type	Ethyl butanoate	Ester	Fruity	[35]
	Ethyl hexanoate	Ester	Fruity	[35]
	2-Heptanone	Methyl ketone	Mouldy flavor, spicy, cinamon	[34]
	2-Nonanone	Methyl ketone	Mouldy flavor, fruity, floral	[34]
	Alkan-2-one	Methyl ketone	Mouldy flavor	[18]

## 2. Novel technologies to improve cheese flavor

### 1) Application of adjunct culture for cheese

Cheese ripening is a complex process to contain microbiological and biochemical events affecting the formation of characteristic cheese flavors. During cheese making, microbial metabolism contributes to creating a unique flavor in cheese [18]. Although starter cultures have been used to reduce cheese pH due to the conversion lactose to lactic acid, non-starter cultures are used to strengthen the flavor of cheese or shorten the ripening period. Non-starter cultures contain different growth profiles compared with starter culture applied in cheese manufacture. Unlike starter culture, bacterial concentration is initially low in a range of  $10^2$  to  $10^3$  CFU/g after curd formation. The number of non-starter bacteria increases to  $\sim 10^8$  CFU/g during cheese ripening [36]. Adjunct cultures are non-starter cultures, which are added to cheese milk for enhancing the formation of cheese flavor during ripening [37]. Unlike naturally developing non-starter culture, adjunct cultures are designedly selected and used for cheese. They are also used to decrease cheese ripening time, which can enable considerable cost

reductions for the cheese industry [37].

Typically, low-fat cheeses, such as low-fat Feta, have less intense flavors than full-fat cheeses. The use of adjunct cultures containing *Lactococcus lactis* and *Lactococcus cremoris* resulted in the development of similar flavors to the full-fat Feta [38]. It was reported that an increase in the formation of critical cheese flavor compounds, such as diacetyl and acetoin, was observed in short ripened Caciotta cheese treated with an adjunct culture, *Lactobacillus paracasei* 4321 compared to control [36]. In Cheddar and Swiss cheese, the important cheese flavor attribute, fruity flavor, was enhanced when adjunct cultures were used with the inclusion of ethanol. An increase in the development of fruity flavor in cheese can be attributed to the production of ethyl ester [39,40]. In Camembert cheese, the formation of ethyl esters was significantly increased when *Lactobacillus plantarum* CCFM 12 containing high esterase and alcohol acyltransferase activity was used as an adjunct culture [40]. The development of esters in Camembert cheese could be due to the ethanol content and enzyme functions involved in the synthesis and decomposition of esters [40].

## 2) Enzymatical approaches for cheese flavor

Enzymes added from an external source are referred to as exogenous enzymes. Exogenous enzymes including lipase have been used to facilitate the development of specific cheese flavor profile and accelerate cheese ripening [18]. Those exogenous lipases are mainly obtained from microbial sources, such as *Lactobacilli* and yeast, due to the simplicity of its manufacture [41]. The use of microbial lipase to Swiss cheese led to an accelerated cheese ripening resulting in a decrease in the cost of cheese manufacture while the physicochemical properties of cheeses was maintained [42].

However, the use of exogenous lipases for enhancing cheese flavors could induce excessive lipolysis resulting in cheese flavor defects [18]. To minimize these issues, the immobilization of lipases receives notable attention since it can increase the activity and stability of lipase and modulate the enzyme-substrate interactions [43,44]. When the lipase is immobilized in delivery systems, such as emulsion and nanoparticle, more free fatty acids could be formed due to an increase in lipase activity and chemical interactions between lipase and substrate [45]. As mentioned earlier, low fat cheeses contain the flavor defects of cheeses compared with full fat cheeses although consumer demands are increasing. In low fat cheeses, a reduced amount of aromatic free fatty acids could be developed by lipases because of lower fat content [46,47]. To enhance the release of aromatic free fatty acids in low fat cheeses, lipases immobilized on lactalbumin nanotubes were employed for stabilizing oil-in-water Pickering emulsion [46]. The addition of oil-in-water Pickering emulsion treated with lipases immobilized on lactalbumin nanotubes to low fat cheeses resulted in the formation of a significantly higher amount of aromatic free fatty acids. It could be attributed to enhanced contact area between fat and immobilized lipase [46]. On the other hand, adding lipases immobilized on liposome to cheese milk significantly increased the development of volatile fatty acids and promoted the lipolysis and ripening of Cheddar cheese [48].

## Conclusion

This review discusses how the metabolism of milk fat contributes to the development of cheese flavor. The hydrolysis of milk fat, facilitated by microbial and endogenous lipases, leads to the formation of fatty acids, including short-chain, medium-chain, and free fatty acids. This chemical process is crucial for the development of cheese flavor compounds during ripening. Two major approaches, such as the application of adjunct culture and the immobilization of lipases, are discussed to minimize the flavor issues of low-fat cheese during ripening. The application of adjunct culture results in an increase in cheese flavor and accelerates the ripening process. The immobilization of lipases leads to the production of cheese flavor compounds and promotes ripening. It is concluded that a microbial approach using adjunct culture and an enzymatical approach including lipase immobilization can be effective ways to enhance cheese flavor during ripening.

## Conflict of Interest

The authors declare no potential conflict of interest.

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