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Unveiling the aroma profile of anhydrous milk fat: Insights into key odorants during biocatalysis by aroma extract dilution analysis, aroma recombination, and omission experiment

Abdelaziz Elbarbary [a,b], Abdullah S. Seddiek [c], Yulong Shao [a], Mingdi Yu [a], Jun Jin [a,*], Youfeng Zhang [a,d,*], Xingguo Wang [a,e]

[a] State Key Laboratory of Food Science and Resources, School of Food Science and Technology, Jiangnan University, Wuxi 214122, China
[b] Dairy Science Department, Faculty of Agriculture, Benha University, Moshtohor, 13736, Egypt
[c] Department of Food Science, Faculty of Agriculture (Saba Basha), Alexandria University, Alexandria 21531, Egypt
[d] Department of Flavor Chemistry, Institute of Food Science and Biotechnology, University of Hohenheim, Fruwirthstr., 12, 70599 Stuttgart, Germany
[e] Food Laboratory of Zhongyuan, Luohe, Henan Province, 462300, China

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ABSTRACT

Determination and verification of the key aroma compounds of anhydrous milk fat (AMF) were conducted during biocatalysis at different times using commercial immobilized Lipozyme RMC from *Rhizomucor miehei* and Lipozyme TL-IM from *Thermomyces lanuginosus*. The analysis techniques employed included headspace-solid phase microextraction-gas chromatography-mass spectrometry (HS-SPME-GC–MS) coupled with olfactory port, aroma extract dilution analysis (AEDA), odor activity values (OAV), aroma recombination model, and omission experiments. A total of 17 main odorants were identified and quantified in AMF, and characterized by flavor dilution factors ranging from 4 to 128. Among them, butanoic acid, methyl octanoate, hexanoic acid, methyl butanoate, and methyl decanoate revealed the highest flavor dilution. These aroma-active compounds were confirmed as important odorants based on their OAVs ≥ 1. Furthermore, the omission and recombination model sensory experiments successfully identified 12 key odorants including butanoic acid, methyl octanoate, hexanoic acid, octanoic acid, methyl hexanoate, and methyl butanoate in AMF catalyzed with Lipozyme RMC. Overall, this research provides valuable insights into the enzymatic modification of AMF and its impact on flavor modulation in dairy products.

1. Introduction

Anhydrous milk fat (AMF), a major fatty product worldwide, contributes to the food sector due to its nutritional content, rich taste, aromatic properties, and health advantages (Bielecka et al., 2023; Karakus et al., 2022). The aroma is a distinguishing characteristic of these fatty products. In recent years, China's dairy industry has undergone a significant transformation in the production of AMF from cream or butter, based on centrifugal separators. However, these AMF products may possess a slight flavor (Battula et al., 2020; Illingworth et al., 2009). Clean-label consumer trends are pushing food companies to switch from synthetic flavors to "cleaner" natural milk fat flavors due to the unique fatty acid composition of milk fat, C4-C12 (Staniewski et al., 2021; Yang et al., 2018). When these fatty acids are synthesized *via* enzymatic processes, they attain the status of "natural flavors" in accordance with the standards established by the International Organization of the Flavor Industry (Gandhi, 1997; Martins et al., 2013).

Lipases (EC 3.1.1.3), are frequently employed in the food processing industry as a hydrolysis reaction method to enhance food products' flavor, texture, and overall taste (Sutay Kocabaş et al., 2022; Zhang et al., 2020). However, in milk fat, the quality of lipolysis (i.e., enzymatic hydrolysis of triacylglycerol) causes free fatty acid to be formed (Sarmah et al., 2018; Zhang et al., 2020), which can change over time, resulting in a rancid flavor in milk fat that directly impacts the dairy industry. Therefore, it is imperative to investigate alterations in substances generated by lipase reactions through the hydrolysis process to

* Corresponding authors.
E-mail addresses: abdelaziz.elbarbary@fagr.bu.edu.eg (A. Elbarbary), abdullahsaad@alexu.edu.eg (A.S. Seddiek), 1012220220@stu.jiangnan.edu.cn (Y. Shao), mingdiyu111@163.com (M. Yu), junjin@jiangnan.edu.cn (J. Jin), youfeng.zhang@uni-hohenheim.de (Y. Zhang), xingguow@jiangnan.edu.cn (X. Wang).

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