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Review paper

Non-thermal technologies for milk and dairy processing

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ARTICLE INFO	A B S T R A C T	
ARTICLE INFO Keywords: Non-thermal technologies Dairy processing Functional foods	Modern consumers demand minimally processed, high-quality, sustainably produced, and ethically sourced food. The food industry strives to meet these demands without compro- mising food safety. Non-thermal technologies offer a solution by using different physi- cal hurdles to ensure microbiological safety and extended shelf life. In the dairy indus- try, high-pressure processing, ultrasound, ultraviolet processing, cold plasma and pulsed electric fields show promise as effective non-thermal technologies. These methods achieve microbial inactivation by altering cell membrane structures and damaging genetic mate- rial, although the specific mechanisms may vary. Moreover, non-thermal technologies have the potential to enhance product quality and facilitate the development of functional dairy products, with high-intensity ultrasound and supercritical carbon dioxide as particularly noteworthy. Despite the expanding research and development in the field of non-thermal technologies in dairy industries, several challenges persist, including equipment costs, en- zyme inactivation efficiency, the absence of validation procedures, regulatory hurdles and consumer acceptance.	

1. Introduction

Modern consumers are becoming more informed and aware of the food they purchase — they search for the food that is minimally processed, has high quality, and a "fresh-like" appeal (*Neokleous et al.*, 2022; *Zhang et al.*, 2018). Additionally, sustainability and ethics have emerged as crucial elements that consumers take into account when choosing food for purchase (*de Toledo Guimarães*, 2018). The food industry strives to meet consumers' demands, but at the same time, food safety must not be compromised, and long shelf-life needs to be achieved (*Mir et al.*, 2016).

Conventional heat treatments are routinely applied in the dairy industry in order to achieve microbiological safety, inactivate enzymes and prolong shelf life, but they also result in loss of nutritional and sensory properties of the product (*Delorme et al.*, 2020). Moreover, the issue of sustainability in milk processing is gaining more attention, due to the high carbon footprint associated with the milk supply chain (*Grandsir et al.*, 2023). Namely, milk is recognized as a second most polluting drink in the world after coffee (*Poore and Nemecek*, 2018).

While the search for new preservation and processing technologies has been ongoing for more than 100 years, global trends in food production have accelerated research in the field of novel food processing technologies. At present, the research focus on novel food-processing is divided into two distinct fields. The first field is focused on novel thermal technologies that utilize heat generated through unconventional ways, such as microwaves and rad-

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Published by Institute of Meat Hygiene and Technology — Belgrade, Serbia This is an open access article under CC BY licence (http://creativecommons.org/licences/by/4.0) iofrequency. The second field involves non-thermal technologies that rely on physical hurdles to achieve desired goals (*Barbosa-Cánovas & Bermúdez-Aguirre*, 2010)

Non-thermal technologies are defined as the technologies which are applied at the ambient or sublethal temperatures (Cullen et al., 2012), without direct exposure of the product to heat (Chacha et al., 2021). Non-thermal technologies rely on the use of electrical, electromagnetic, light or mechanical forces instead of thermal energy (Rodriguez-Gonzales et al., 2015). Since thermisation is the mildest heat treatment applied in the dairy industry (57-68°C for 10-20s), in this context, processing milk at temperatures below 57°C can be considered as non-thermal treatment (Scudino et al., 2020). However, it should be taken into consideration that EU legislation defines milk as raw only if it has not been heated at the temperatures above 40°C, or exposed to any other treatments with an equivalent effect (EFSA, 2015).

The most promising and most investigated non-thermal technologies in the dairy industry are high-pressure processing (HPP), ultrasound, ultraviolet processing, cold plasma and pulsed electric fields (PEF).

The aim of this review is to give brief overview of these technologies, their potential for the future application, but also current limitations. Membrane filtration, as one of the most important non-thermal technologies, is not covered by this review, since it is already widely used in the dairy industry.

2. Application of non-thermal technologies in the dairy industry

Initially, non-thermal technologies were meant to be employed to ensure food safety. However, other potential uses also emerged over time. An overview of the non-thermal technologies which have the potential for application in the dairy industry, mechanisms through which they achieve microbial inactivation and other potential uses are shown in Table 1.

Microbial inactivation by non-thermal technologies is accomplished through two mechanisms: alteration of the cell membrane structure and destruction of the genetic material (*Zhang et al.*, 2018). However, the specific ways in which these effects are achieved may vary among different non-thermal technologies (Table 1). Microbial inactivation depends on many factors, such as cell shape and size, initial number and phase of growth, but also properties of the food matrix and process variables (Abrahamsen & Narvhus, 2022; Soltanzadeh et al., 2020). Due to the limited effectiveness of individual non-thermal technologies in microbial inactivation, the focus has shifted towards exploring the potential of combining different non-thermal technologies or integrating non-thermal technologies with heat treatments. This approach aims to achieve the desired level of microbial inactivation by synergistic effects (Evrendilek, 2014). Attention has been paid to studying the impact of combining ultrasound with heat, known as "thermosonication"; the effects of combining ultrasound, heat, and pressure, referred to as "manothermosonication", as well as the combination of ultrasound with ultraviolet irradiation, known as "photosonication" (Abrahamsen & Narvhus, 2022). The combination of pulsed electric field (PEF) and mild heat treatments has demonstrated promising results, surpassing the microbicidal effects achieved by either PEF or heat pasteurization alone (Sharma et al., 2014; Alirezalu et al., 2020). This combined approach offers microbial safety, but at the same time, superior quality in terms of nutritional profile and sensory attributes (Soltanzadeh et al., 2020).

Additional advantages of non-thermal technologies were recognized, such as the potential to improve product quality and to help the development of novel food products (Barbosa-Cánovas & Bermúdez-Aguirre, 2010). Several non-thermal technologies are being extensively researched for their potential for the production of functional dairy products. High-intensity ultrasound and supercritical carbon dioxide have emerged as the most promising ones (de Toledo Guimaraes et al., 2018). Ultrasound treatment has proven to be effective in achieving a fermented milk product with increased levels of biologically active compounds (Potoroko et al., 2018). It has also demonstrated its ability to improve lactose absorption by bifidobacteria and promote the production of organic acids (Nguyen et al., 2012), and enhance the viability of the probiotic strain Lactobacillus acidophilus-La5 (Barukčić et al., 2015). Additionally, it has been utilized to improve the antioxidant and antihypertensive activities, as well as the bioavailability of phenolic compounds and bioactive peptides in chocolate milk beverages (Monteiro et al., 2018). Lastly, ultrasound treatment has been employed in the manufacturing of y-aminobutyric acid (GABA)-enriched products (Shokri et al., 2021).

Process	Definition	Mechanism of microbial inactivation	Other potential uses, use/ beneficial effect
High pressure processing	Transfer of high hydrostatic pressure (in the range 100–1000 MPa) to the food matrix, using a medium (water or oil-alcohol mixtures) (<i>D'Incecco</i> <i>et al.</i> , 2021)	Under the influence of high pres- sure, the cell wall and cell mem- brane of microorganisms undergo irreversible destruction, resulting in a notable change in their per- meability. Secondary, tertiary and quaternary structures of large molecules ae changed and these molecules lose their function (<i>Zhang et al.</i> , 2018).	Cheese ripening. Enhancing sensory properties of yogurt (texture and cream- iness). Prevention of post-acidifica- tion due to inactivation of start- er cultures, yeasts and moulds. Manufacture of reduced-fat and stabilizer-free ice cream (<i>Voigt et al.</i> , 2015). Improvement of functional properties (<i>de Toledo Guima- rães et al.</i> , 2018).
Ultrasound	Longitudinal sound waves, occur- ring at frequencies beyond the hu- man hearing threshold (approximate- ly 20kHz). Ultrasound creates a se- quence of compressions and expan- sions within a medium (food matrix). This process triggers the formation of vacuum bubbles, which subsequently grow and collapse (<i>D'Incecco et al.</i> , 2021).	Micro bubbles in milk generat- ed during the treatment under- go expansion and contraction, in the process known as cavitation. These bubbles gradually increase in size until they collapse, result- ing in the generation of localized high temperature and pressure. In such conditions, the bacterial cell membranes are ruptured and de- stroyed. Chemical compounds that break down the cell walls are also formed (<i>Abrahamsen & Narvhus</i> , 2022).	Separation and extraction of milk fat. Homogenization Improvement of lactose crys- tallization in ice-cream. Improvement of emulsifying properties of dairy emulsion. Reduction of viscosity and control of age thickening in concentrated milk (<i>Abraham-</i> <i>sen & Narvhus</i> , 2022).
Ultraviolet radiation	Non-ionizing form of invisible light in the portion of electromagnet- ic spectrum between visible light and X-rays, with the wavelength be- tween 100 and 400 nm (<i>Delorme et al.</i> , 2020).	Formation of photoproducts, which interfere with DNA transcription and replication processes (<i>Delorme et al.</i> , 2020).	
Cold plasma	Plasma, an ionized gas considered the fourth state of matter, compris- es electrons and positive and nega- tive ions, which can transfer ener- gy through collisions with gas mol- ecules (gas is in a quasi-neutral state due to the existence of equal number of positive and negative charges car- ried by different species) (<i>Neoκleous</i> <i>et al.</i> , 2022). Consequently, this pro- cess generates highly reactive spe- cies like reactive hydroxyl radicals, hydrogen peroxide, ozone, nitrogen oxide and UV radiation. (<i>Niemira</i> , 2012).		Enhancement of proteins' physical and chemical proper- ties (<i>Jadhav et al.</i> , 2021).
Pulsed electric field (PEF)	Passage of an alternating current us- ing electrodes. Food matrix is placed between the electrodes, and is treated with short high-voltage electric puls- es, generated by high-voltage pulse generator (<i>D'Incecco et al.</i> , 2021).	Electric field generated by PEF inactivates microorganisms through the process of electropo- ration (<i>Alirezalu et al.</i> , 2020).	

Table 1. Non-thermal technologies for milk processing and their potential use in the dairy industry

3. Non-thermal technologies in dairy industry — considerations for future use

Despite the promising and expanding research and development in the field of non-thermal technologies in dairy industries, numerous challenges still remain to be addressed.

One of the current limitations of non-thermal technologies for industrial use is the requirement for significant initial investments, primarily due to the high cost of equipment. As a result, these technologies are more suitable for premium-priced products rather than mass-market applications (*Voigt et al.*, 2015; *D'Incecco et al.*, 2021). Furthermore, the existing equipment is designed for in-batch processing, thereby limiting the amount of product that can be processed at one time (*Voigt et al.*, 2015; *Arshad et al.*, 2020). For industrial implementation, it is necessary to develop equipment that can operate continuously in-line with the production process (*D'Incecco et al.*, 2021).

In order for the process to be considered a valid alternative to heat-treatments, it is crucial that the native or bacterial enzymes in milk can be effectively inactivated. Various non-thermal technologies, including ultrasound, HPP, and PEF, have demonstrated effectiveness in enzyme inactivation. However, when used individually, these methods require high-intensity treatment parameters, which can have detrimental effects on the nutritional and sensory properties of milk. A combination of different non-thermal technologies or the integration of non-thermal technologies with mild heat treatments can be employed, in order to achieve desired results (*Ahmad et al.*, 2019).

Effects of non-thermal technologies on sensory properties of the products have not been studied extensively to date (*Neokleous et al.*, 2022). Some studies reported possible negative impacts of non-thermal technologies on flavour and aroma of the products, mainly due to the oxidation processes (*Priyadarshini et al.*, 2019; *Choudhary & Bandla*, 2012; *Delorme et al.*, 2020; *Ribeiro et al.*, 2021; *Marchesini et al.*, 2015).

The efficacy of high temperature-short time pasteurization is typically evaluated by the activity of alkaline phosphatase, with remaining activity indicating an inadequate process. However, for non-thermal technologies, standardized process parameters and validation procedures have not yet been established. Consequently, the absence of validation procedures, but also harmonized labelling, represent regulatory hurdles for the non-thermal technologies (*Barbosa-Cánovas & Bermúdez-Aguirre*, 2010; *D'Incecco et al.*, 2020).

Consumer acceptance of novel products remains a challenge (*Tuorilla & Hartmann*, 2020), as there is a perception that these products are overpriced and pose a higher risk compared to conventionally processed alternatives (*Coutinho et al.*, 2021). It is crucial, therefore, to educate consumers about the potential advantages of non-thermal technologies to achieve better acceptance (*Coutinho et al.*, 2021).

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