Non-thermal Treatment of Milk: Principles and Purpose

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Introduction

Milk is a natural secretion from healthy mammals that contains nutrients such as fat, protein, lactose, minerals and vitamins, and represents an excellent dietary source of both major and minor nutrients. Milk has been consumed routinely for decades by all age-groups (neonates to elderly) to meet their nutritional requirements throughout life.

Although healthy animals produce almost sterile milk, post-milking contamination takes place from external factors such as environment, machines, pipelines, containers and persons that come into contact during milking, storage and transportation. Unfortunately, milk then acts as an excellent growth medium for most microorganisms. In particular, in the absence of preservation, spoilage and pathogenic microorganisms proliferate in milk, limiting its shelf-life and leaving it potentially unfit for human consumption. Hence, this highly perishable material needs timely processing to retain its quality attributes.

Different classical (such as thermization, low and high temperature pasteurization, in-bottle sterilization, ultra-high temperature treatment and ultra-pasteurization) and novel (e.g., ohmic heating, microwave heating, radiofrequency heating) processes have been used to treat milk with the goal of rendering it safe for human consumption. These processes also extend the shelf-life of milk by destroying spoilage microorganisms and inactivating enzymes.

Traditional and novel heat preservation methods are known to induce changes in milk components that could be desirable as well as undesirable. For example, heating of milk at higher temperature (>100 °C or more) will decrease its nutritional quality by decreasing vitamin contents, inducing denaturation of milk proteins and imparting brown color and cooked flavor. The intensity of such adverse changes in milk is a function of applied processing time-temperature combinations, and the extent of these adverse changes could be decreased/limited to some extent by novel thermal processes but cannot be omitted completely.

Consumers are now very well aware and concerned about the quality of food which they eat. Hence, they are now demanding milk and other food products that retain fresh-like sensory attributes, maximum wholesomeness and high levels of nutrients due to minimal processing, and that are safe to consume (i.e., free from microorganisms and chemical additives). Therefore, convenient, safe, nutritive, fresh and long-life foods can be considered as a right of consumers now.

Non-thermal processing of milk and other foods includes their processing at ambient temperatures for shorter durations to retain better quality and sensory attributes and higher retention of nutritional components, and at the same time to retain or even improve safety aspects. These products are also free from chemical additives. Thus, non-thermal treatments can potentially ensure food safety and shelf-life stability in milk and other food products in a superior, economic and sustainable way.

Non-thermal treatments like high pressure (HP) treatment, pulsed electric fields (PEF), high-pressure homogenization and microfluidization, ultrasound (US) and pulsed light technology (PLT) are capable of enhancing the shelf-life of treated food products with improved product quality and safety. This article provides an overview of the principles and purposes of these treatments.

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High-Pressure Processing (HPP) Technology

HPP technology is also known as High-Pressure (HP) Treatment, High Hydrostatic Pressure Processing (HHPP), Pascalization and Ultra HighPressure Processing (UHPP). Broadly, the rationale of the HPP process can been explained based on two main principles, namely Le Chatelier's principle and Pascal's principle, or the isostatic principle. Arora and Cauhan (2019), Martinez-Monteagudo and Balasubramaniam (2016) reported that there are actually four principles which govern the effects of HHP on foods; the additional principles are the microscopic ordering principle and the Arrhenius relationship.

Le Chatelier'sPrinciple states that "if a system in equilibrium is imposed to change its conditions then the system will restore the equilibrium by counteracting the imposed change". This principle is based on the second law of thermodynamics and is valid for reversible processes. This principle explains the effects of pressure and temperature on physical, chemical and biological phenomena. According to Le Chatelier'sPrinciple, any reaction of phase transition phenomenon accompanied by a reduction in volume is favored by high pressure, whereas those leading to volume expansion would be inhibited (Singh et al., 2019). Pascal's principle states that "pressure can be instantaneously and uniformly applied throughout the sample as HPP is a volume-independent process without any pressure gradient" (Voigt et al., 2015). The microscopic ordering principle states that, for a given substance, the degree of molecular ordering is enhanced with increasing pressure at a fixed temperature (Martinez-Monteagudo and Balasubramaniam, 2016).

Upon HP treatment of milk at 100, 300 and 500 MPa, water is compressed by 4%, 10% and 15%, respectively (Hinrichs et al., 1996; Huppertz, 2010). Such compression of water is also accompanied by an increase in temperature of the milk at the rate of 2–3 °C per 100 MPa rise in applied pressure (which is known as adiabatic heating).

Key Parts of an HPP System

The most vital part of any HHP system is its pressure vessel. Liquid samples may be treated as such (without packaging) while other samples in their pre-packed form are placed in the pressure vessel. In order to process the pre-packed foods, a pressure transmitting medium is required. Different pressure-transmitting liquids such as water (most common), water-oil emulsions, sodium-benzoate, ethanol and food grade water-ethanol solutions, silicon and caster oils (Balaubramaniam et al., 2015) are available and their selection and use is governed by their heat of compression, corrosion-resistance properties, sealing ability under pressure, and changes in their viscosity under pressure (Arora and Cauhan, 2019). The surfaces of the cylindrical pressure vessel which are in direct content with food material is either made of corrosion-resistant material (like stainless steel) or adequately protected with the addition of anti-corrosive agents in the pressure-transmitting liquid.

Fig. 1 shows the main parts of an HPP system, which includes the pressure vessel and its closure; high pressure pumps or intensifiers (to generate required pressure); a medium to transfer generated high-pressure; and devices to measure and control level, temperature, pressure and flow of material, along with a robust material handling system.

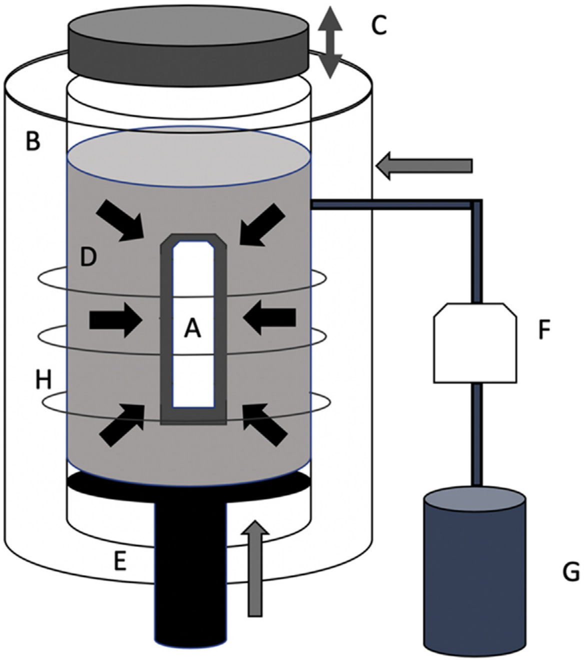

Figure 1 Schematic figure of a high-pressure (HP) treatment system for food. A food product to be treated, in a suitable package (A) is placed in a cylindrical chamber with thick metal walls (B). This chamber is sealed by a suitable closure (C), filled with a pressure-transmitting medium (D) and pressure is generated uniformly (*dark arrows*), either through direct compression of the medium using a piston (E) or through a high-pressure pump (F) pumping medium from a reservoir (G). Temperature may be controlled during treatment by a suitable heat-exchange system (H).

Pressure vessels could be monoblock (1 piece, suitable for <400 MPa pressure) or may comprise two or more concentric cylinders which are available in different volumes suitable for lab-scale (0.1–2 L), pilot plant (10–25 L), batch and commercial production facilities (>100 L); these can be placed horizontally (easy and fast loading and unloading), vertically (time-consuming and difficult in material handling) and even in tilting positions for easy handling/loading and unloading of food materials. After loading, both ends of the cylindrical vessel are covered by closures and pressure is applied on their external surface; a yoke is used to restrain end closures while under high axial pressure (Arora and Cauhan, 2019). Apart from these, a pressure creating device is another important component of the HHP unit; a low-pressure pump is generally used with lab and pilot plant units to create fast, direct piston-type compressions, while an intensifier is used in large scale plants for indirect compression to create very high pressures. To achieve the desired pressure, the extent of compression is governed by the compressibility of working fluid and product. Moreover, the compression time and horsepower of the pump are directly related. Food packages should be free from gas and empty spaces for best pressure transmission in HPP. The success of this technology demands a uniform increase and control of temperature. More detailed information on laboratory, pilot and commercial scale HPP plants can be found in Ref. Balasubramaniam et al. (2016).

HP Treatment of Milk

HPP experiments on milk were likely first conducted in 1899 at the West Virginia Experimental Station by Hite (1899), who reported a marked increase in shelf-life of the milk after treatment with HPP. Furthermore, after HPP treatment, milk become more translucent, indicating that HPP induced some changes in its physical properties.

The major issues associated with thermal processing of milk including loss of nutrients, browning, and loss of bioactive compounds can be mitigated by HPP. Application of HPP at ambient conditions needs less energy, decreases operational costs and increases production sustainability (Singh et al., 2019). HPP treatment of milk (>400 MPa) leads to the inactivation of pathogenic and spoilage microorganisms by different mechanisms such as inhibition of protein synthesis, decreased number of ribosomes, leaking of cellular components, denaturation of critical enzymes and alteration in permeability of cell membrane. For longer treatments, HPP can also separate cell walls and cell membranes, inducing elongation in cells and restricting their ability to move. Pressure-induced conformational changes in functional proteins alter their cell division (Singh et al., 2019).

Generally, pressures >200 MPa at ambient temperature are needed to induce inactivation of bacterial cells. Treatment at 400–600 MPa at ambient temperature leads to the destruction of the vegetative cells of bacteria, yeast, and fungi; however, pressure-resistant strains may still survive (Singh et al., 2019). Efficacy of HP treatment in terms of microbial destruction depends on different factors related to characteristics of the food and the bacteria, processing conditions, and physiological state of microorganisms. Gram-positive and gram-negative microorganisms can be inactivated by treatment at 500–600 MPa and 300–400 MPa for 10 min at 25 °C, respectively. The presence of teichoic acid in the cell wall of gram-positive microorganisms offers better structural integrity and rigidity to their cell wall, hence the higher pressures needed to inactivate these organisms (Singh et al., 2019). The baro-resistance of yeast and molds is lower than that of bacteria, and their vegetative forms are most pressure-sensitive compared to their spores.

Processing of milk at 400 MPa for 15 min or at 600 MPa for 3 min could produce a product comparable to pasteurized milk by effectively destroying pathogenic and spoilage microorganisms without the need for heat treatment. To produce sterilized products employing HPP alone, even application of very high pressures (>1000 MPa) is not sufficient to inactivate bacterial spores, but, if these spores are allowed to germinate at 50–300 MPa, subsequent application of mild pressure or heat treatment may inactivate them (Smelt, 1998).

Recently, Stratakos et al. (2019) studied the effect of HP treatment on safety, shelf life and quality of raw milk by externally inoculating *Escherichia coli*, *Salmonella* and *L. monocytogenes* (pathogens) into the milk followed by treatment at 400–600 MPa for 1–5 min, which decreased bacterial levels by 5 log CFU/mL.In addition to safety aspects, HP-treated milk has a better shelf life than pasteurized milk (Stratakos et al., 2019).

Generally, the basic purpose of HP pasteurization of milk includes the elimination of pathogens, shelf-life enhancement, and production of innovative, fresh and clean-label products. Today, some HPP-treated milk products are commercially available, such as the "Made by cow" brand in Australia, which is described as cold-pressed raw milk, and which is sold unhomogenized in glass bottles. It is possible that such products will emerge in other countries, reflecting consumer demand for more natural less-processed products, but in each case will have to be certified as safe for sale by appropriate regulatory bodies.

Effect of HP Treatment on Milk Constituents

HP treatment has significant effects on the constituents of milk, some of which are similar to those of heat treatment (e.g., denaturation of β-lactoglobulin) and some of which are unique to HP treatment compared to other physical processes (e.g., aggregation or dissociation of casein micelles); in addition, many heat-induced changes in milk, such as Maillard reactions, do not occur on HP treatment, while phenomena such as enzyme inactivation differ in a case-specific manner. High-pressure induced changes in milk constituents and milk attributes are summarized in **Table 1** and were recently reviewed by Singh et al. (2019).

Table 1 Effect of HP treatment on milk constituents and attributes

Effect on milk constituents	
Minerals	• Reversible solubilization of micellar calcium phosphate (MCP) up to 400 MPa.
Fat	• HP treatment of cream (100–400 MPa) induced crystallization (maximum at 200 MPa) in milk fat in cream. Cream treatment at 800 MPa for 10 min increased fat globule size. • Treatment at $\leq$200 MPa increased crystallization and melting temperatures of milk fat by 16.31 °C and 15.51 °C per 100 MPa pressure. • $\leq$500 MPa: Modified size and distribution of milk fat globules. • Association of β-lactoglobulin, κ-casein and α-lactalbumin with the MFGM in milk HP-treated at $\geq$100, $\geq$500 and $\geq$700 MPa, respectively. • Decreased whipping time and serum loss in cream treated at 500–600 MPa for 1–2 min. • Milk treated at 100–250 MPa showed higher creaming and cold agglutination than control sample; however, treatment at $>$400 MPa reduced cold agglutination and creaming.
Lactose	• No Maillard reaction. • HP treatment of milk at 100–400 MPa, for 10–60 min at 25 °C: no changes.
Casein	• Milk treatment at 400 MPa lead to micellar disruption and solubilization of MCP. Increases in pressure, temperature and decrease in pH further increases it. The size of casein micelles decreases in HPP treated milk as a function of applied pressure. However, this change in size may be reversed back during storage of milk. • Such changes in casein micelle size also changes turbidity and lightness of skimmed milk, with little effect at 100–200 MPa, but greater decreases with increases in applied pressure.
Whey protein	• Milk treated at $\leq$100 and $\geq$400 MPa, caused no and 90% denaturation of β-lactoglobulin (β-lg), respectively, while denaturation of α-lactalbumin (α-la) starts at $\geq$400 MPa and reaches about 70% after its 30 min treatment at 800 MPa. Extent of denaturation was governed by treatment time, temperature, pH and level of MCP. • Milk treated at $\leq$100 and $\geq$400 MPa showed no denaturation of bovine serum albumin; treatment at 500 MPa for 5 min caused only 10% denaturation of colostral immunoglobulin G.
Enzymes	• HP treatment of milk at $\leq$400 MPa did not inactive lipoprotein lipase, xanthine, oxidase and lactoperoxidase, and decreased shelf-life of milk due to enhanced lipolysis. • Phosphohexose isomerase, γ-glutamyl transferase and alkaline phosphatase were completely inactivated in milk treated between 550 and 800 MPa.
Water	• HPP treatment of milk at 100, 300, 500 and 600 MPa, caused 4%, 10%, 15%, 15% compression of water.
Effect on milk attributes	
Color	• HP-treated milk possesses similar color values quality compared to raw milk.
Flavor	• Beside flavor compounds, vitamins and amino acids remain unaffected in HPP treated milk.
pH and freezing point depression	• One unit decrease in pH of milk at every 1000 MPa pressure. Milk freezing point was decreased to -8 and -22 °C at 100 and 210 MPa.
Viscosity	• HP treatment increased viscosity of treated sample due to disruption of casein micelles and denaturation of whey proteins. • Concentration of milk and treatment at high pressure further increases it. St more than $>$300 g/kg total solids and $>$400 MPa, pressure strongly enhances viscosity and even cause gelation of milk in extreme conditions.

Arora and Chauhan (2019), Balasubramaniam et al. (2015, 2016), Deeth et al. (2013), Voigt et al. (2015), and Singh et al. (2019).

High-Pressure Homogenization (HPH) and Microfluidization

The dairy industry has been using conventional homogenization (10–30 MPa) for at least a century to prevent the formation of a cream layer at the milk surface during storage and also to facilitate the manufacture of dairy products with improved flavor, texture, taste and shelf-life. Homogenizers which can operate in the pressure range 100–300 MPa (even 400 MPa in some cases) are now available and being used for research and industrial applications. Such homogenizers are known as high-pressure homogenizers or ultra-high-pressure homogenizers and micro-fluidizers (Huppertz, 2011a,b; Tobin et al., 2015). "Radial diffuser", "counter-jet-disperser", and "axial-flow-nozzle" are types of high-pressure homogenizers.

High-pressure homogenizers typically consist of a piston valve and a valve seat that is made of ceramics. Ceramics can withstand high pressures and high-stress conditions encountered in this high-pressure homogenization process. The working principle of this process includes the creation of required (high) pressure by a high-pressure pump, followed by the passage of pressurized product through the narrow valve gap existing between valve seat and piston valve and further discharge of product from the valve at atmospheric pressure. The high potential energy of the product is converted into kinetic energy after its passage through a narrow gap.

During this process, milk particles experience different simultaneous and interlinked physical effects such as shearing, high velocity, collision, cavitation, turbulence and sudden increases and decreases in pressure that lead to disruption of milk droplets. Fat globules in the milk droplets are also impacted by high implosive forces during the cavitation (pressure drop) phenomenon of this process.

Pressure intensity is regulated by adjusting the valve gap, which also determines the magnitude of pressure and velocity encountered by the liquid in the homogenizing valve. Very high fluid velocities (100–200 m/s) result in extremely short residence time of milk in this process. Almost linear (0.15–0.2 °C/MPa) increases in milk temperature are associated with an increase in homogenization pressure. The increase in temperature depends on the fat content of milk (0.5 °C/% fat at 150 MPa).

Microfluidization technology is based on a double-acting intensifier pump and an interaction chamber (Huppertz, 2011a,b). A target liquid is forced into the interaction chamber at the required pressure that is generated by air or electric-hydraulic-driven intensifier pump. The target is then divided into two streams as a function of interaction chamber geometry and forced to collide in the reaction chamber at 180 angle, which leads to the formation of an emulsion as a result of subsequent cavitation (sudden pressure drop), shearing and turbulence in the reaction chamber. Pressures up to 300 MPa can be achieved using this technology. An oil phase and aqueous phase or milk (existing emulsion) could be converted into very narrow particle size distribution containing another emulsion employing microfluidization technology. Such a process can result in narrow particle size distribution in milk and cream containing small (>30 mm) fat globules (Huppertz, 2011a,b).

HPH and Microfluidization Treatment of Milk

Several studies have established that milk treated by HPH (>150 MPa and >40 °C) has achieved microbial destruction similar to that achieved by HTST pasteurization processes and aimed for market milk (Hayes et al., 2005; Pereda et al., 2007; Picart et al., 2006; Smiddy et al., 2007). Thus, HPH can serve as an alternative process to conventional pasteurization and homogenization of milk. Furthermore, the shelf-life of HPH-treated milk and conventionally pasteurized and homogenized milk were similar during refrigerated storage (Pereda et al., 2006; Smiddy et al., 2007). HPH treatment of milk may also be more effective than traditional processes in decreasing the extent of creaming in the treated milk because HPH leads to a greater reduction in milk fat globule size (Hayes et al., 2005).

Apart from these positive effects, HPH treatment of milk had some adverse effects, such as the generation of an adverse flavor profile. For example, residual lipase activity in HPH-treated (200 MPa) milk showed higher lipolysis (lower pH and liberation of free fatty acids) potential leading to rancid flavor than that observed with conventional homogenization (Hayes et al., 2005; Pereda et al., 2008). HPH-treated (300 MPa) milk was found to be highly susceptible to lipid oxidation (Pereda et al., 2008). Microfluidization reduced the extent of fat separation during storage of UHT milk wherein conventionally homogenized milk samples were stable for 2–3 months only, while microfluidized milk samples were stable up to 9 months without excessive fat separation (Hardham et al., 2000; Tobin et al., 2015). Various effects of HPH and microfluidization on milk constituents are summarized in **Table 2**.

Table 2 Effect of HPH and microfluidization treatment on milk constituents

Effect on milk constituents	
Milk fat globules	• HPH can decrease size of milk fat globules to a higher extent than that achieved by conventional homogenization. D [3, 2] and D [4, 3] values of raw, conventionally homogenized and HPH (100 MPa) treated milks were 1.0, 0.5, 0.2 and 4.5, 1.0, 0.5 μm, respectively. • Microfluidization is better (optimal pressure 50 MPa) in disrupting milk fat globules than conventional homogenization even at lower pressure.
Proteins	• HPH of milk at ~200 MPa results in partial disruption of casein micelles (could aggregate in presence of calcium at same pressure), but micelle size increases at treatment >250 MPa. • HPH treatment of defatted milk at 30 °C at 300 MPa and 30 MPa applied pressures in first and second stages caused 45% and 30% denaturation of β-lactoglobulin and α-lactalbumin, respectively. • Microparticulated milk proteins are obtained by microfluidization of milk and act as fat replacers. Optimized levels of pressure, temperature and number of passes could produce protein aggregates of desired size and shape. • Microfluidization and heating could produce denatured micro-aggregates of whey proteins with higher heat stability (no sedimentation in heated beverages), and improved foaming and emulsification (better protein absorption) properties compared to non-denatured microfluidized proteins.
Enzymes	• Homogenization of milk at 200 MPa and >40 °C inlet temperature or HPH at 100–200 MPa does not completely inactivate lipase. Applied pressure facilitates lipolysis (decreases pH and can lead to rancid flavor) in milk. HPH-treated skim milk showed no reduction in activity of indigenous plasmin while increases in pressure, fat percent and temperature increased inactivation of this enzyme. • HPH treatment of milk at 200 MPa pressure and 50 °C temperature resulted in significant (<20%) inactivation of plasmin.
Microorganisms	• All physical effects observed by milk during its HPH treatment contributes in inactivation of the wide range of bacterial species. In addition, increase in temperature, pressure and fat content also enhance bacterial inactivation in HPH treatment. • HPH-induced inactivation was higher for gram negative bacteria than gram positive bacteria. Viscosity of the medium is crucial in microbial inactivation in HPH process rather than its (medium) composition. So far, scientific information on microbial inactivation by microfluidization in milk streams is limited.

Deeth et al. (2013), Hardham et al. (2000), Hayes et al. (2005), Pereda et al. (2006), Picart et al. (2006), Stratakos et al. (2019), and Tobin et al. (2015).

Pulsed Electric Field (PEF) Treatment

In Pulsed Electric Field (PEF) processing, short electrical pulses are used to destroy microorganisms and inactivate enzymes present in different foods. High-quality foods can be produced by this method because it induces minimum adverse changes in food components, sensory attributes and physicochemical properties of treated food. Fetterman (1928) patented a process called "Electropure" in which pasteurization of milk was targeted by destroying *Mycobacterium tuberculosis* and *Escherichia coli* through the passage of electric current.

More research has been now conducted on PEFand has elucidated the mechanisms involved in microbial destruction and enzyme inactivation, as well as optimization of process parameters to obtain better retention of nutrients and desired sensory attributes in treated products with enhanced product safety (Upadhyay et al., 2019).

Principles of Pulsed Electric Field (PEF) Treatment

The main principle of PEF technology involves the application of short pulses (1–10 μs) of high electric fields (generated by a high voltage [5–20 kV] pulse generator) to a food material placed inside a treatment chamber between two electrodes, installed with a 0.1–1.0 cm treatment gap and separated by an insulator (Sampedro and Rodrigo, 2015). The duration of these pulses varies between microseconds and milliseconds, while the electric field intensity ranges between 10 and 80 kV/cm (Upadhyay et al., 2019). Applied short pulses (μs) of high voltage leads to the generation of an electric field, which causes inactivation of microorganisms present in food, due to temporary or permanent permeabilization of their (microbial) cell membranes. Bench and pilot-scale PEF plants are now available worldwide which are capable of generating a uniform electric field.

PEF Treatment of Milk and Dairy Products

PEF treatment of milk at 100–550 kJ/L resulted in a 2–5 log10 reduction in different innoculated pathogens, including *Staphylococcus aureus, Escherichia coli, Listeria monocytogenes, Pseudomonas fluorescens* and *Listeria innocua*. Compared to normal heat treatments (72–75 °C, 30–15 s), PEF (~35 kV/cm, 40 °C) had no-significant effect on pH, acidity, free fatty acids, and mineral contents of milk, but decreased the size of the casein micelles and viscosity (Michalac et al., 2003; Floury et al., 2006). Raw, PEF (15–30 kV/cm, 40 °C) treated and pasteurized (75 °C, 15 s) milk showed non-significant differences in their aroma, thiamine, riboflavin, cholecalciferol, tocopherol lactones, acids, or alcohols contents (Bendicho et al., 2002). PEF can possibly act as an alternative to pasteurization; however, their combined use led to higher microbial destruction and higher shelf life owing to their synergistic effect (Sampedro and Rodrigo, 2015). In the future, PEF could be used to improve product quality to meet consumers' demand for fresh-like food products, including dairy products.

Ultrasonication

Principle

Power ultrasound (*f*: 20 KHz to ~1 MHz) is used in food processing and preservation. Ultrasonication treatments are of two main types, i.e., low power (~1 W/cm^2) at high frequency (0.1–20 MHz) and high power (10–1000 W/cm^2) at low frequency (18–100 kHz). Energy-wize, the former conditions do not change food systems, and hence are more suitable for imaging and diagnostic applications, while the latter conditions are used in food processing (the key frequency range is 20–40 KHz, which leads to the most powerful cavitation). Sound waves are non-toxic, environmentally friendly and safe; hence, the application of ultrasound in food processing and preservation has an extra advantage over other techniques (Kentish and Ashokkumar, 2011).

The cavitation induced by ultrasound is the key phenomenon responsible for physical changes in treated food. Ultrasound also generates high localized pressures (up to 100 MPa) and temperatures (up to 5000 K). The bactericidal effect of high power, low-frequency ultrasound is also associated with cavitation. Mechanical vibration, cavitation and, microstreaming are three main physical phenomena of lower frequency ultrasonication (for example, <100 kHz); however, during high frequency (100 kHz–1 MHz) ultrasound treatment, generation of free radicals is an important chemical phenomenon. Free radicals thus formed are responsible for oxidation, and hydroxylation of food in the presence of oxygen can cause chemical changes in food, when oxygen is available; such changes could be beneficial or detrimental (Ashokkumar et al., 2008).

Equipment Setup

Ultrasonication unit mainly consists of the power supply, frequency generator, transducer, and a horn also called sonotrode. Basically, the vibrational energy of a frequency generator is transferred to a horn by a transducer; this horn or sonotrode subsequently transmits these vibrations to the material with which it is in physical contact. The power of lab-scale ultrasonic equipment ranges between 100 and 1000 W, while industrial equipment can generate a power of several kilowatts. Such equipment can even use units in series or parallel configurations to achieve higher treatment capacity (Deeth and Datta, 2011).

Table 3 Advantages, disadvantages and possible applications of selected non-thermal technologies in dairy industry

Non-thermal technology	*Advantages*	*Disadvantages*	*Possible applications*
Hydrostatic pressure processing (Balasubramaniam et al., 2015; Voigt et al., 2015; Trujillo et al., 2016; Singh et al., 2019)	• Generation of high-pressure is rapid while, its distribution throughout samples is quasi-instantaneous uniform. • Minimal or reduced thermal exposure due to instant increase and decrease in temperature during pressurization and depressurization of samples. • Suitable for high moisture–content (liquid and pumpable) foods. • Process efficacy is independent of product shape and size. • Opportunity for novel product formulation as high pressure causes protein denaturation, carbohydrate gelatinization, and fat crystallization. • Pressure accelerates microbial inactivation. • Better consumer acceptance as it is a physical process.	• Batch or semi-continuous operation. • Not suitable for products containing dissimilar compressibility materials. • Throughput limited due to batch operation. • Only pressure cannot fully inactivate bacterial spores and efficacy in enzyme inactivation is also variable. • Higher processing costs and batch operations are barriers for commodity product processing.	• Homogenization and pasteurization of milks. • Treatment of cheese milk (to decrease microbial counts, increase cheese yield and to modify cheese ripening etc.). • Normal and probiotic yoghurt manufacture (less syneresis during storage). • Reduction of whey protein allergenicity. • Ice-cream manufacture (better mouthfeel and creaminess). • Colostrum preservation.
High-Pressure Homogenization (HPH) and microfluidization (Deeth and Dutta, 2011; Huppertz, 2011a,b; Tobin et al., 2015)	• Efficient in milk reductions in fat globule size. • Potential method for homogenization of milk. • Quick, effective and continuous mechanical process. • Lab scale results can reproduce with industrial setup. • Pressure can be changed; uniform particle size reduction. • Interaction chamber of microfluidizer had no moving parts, hence needs less maintenance.	• HPH and microfluidization are expensive, hence, cost effectiveness is a key issue so far. They must be used for the production of high end/high value products. • High energy consumption. • Significant rise in sample temperature during processing. • Unsuitable for industrial scale. • Over processing at high processing pressures. • Development of off flavor due to lipid oxidation and lipolysis.	• Alternative to traditional pasteurization and homogenization of milk. • Pretreatment of milk for manufacture of cheese. • Tailoring functional properties of whey proteins, for example, for improved foaming.
Pulsed electric field (Deeth and Dutta, 2011; Sampedro and Rodrigo, 2015; Upadhyay et al., 2019)	• Suitable for pasteurization of milk, juice, yoghurt, liquid eggs and soups. • Continuous process • Inactivates large number of vegetative bacteria and some enzymes • Along with some thermal treatment, PEF can produce extended shelf life (ESL) milk	• PEF achieves only limited inactivation of bacterial spore and enzymes. • PEF treatment can not destroy bacterial spores. • Commercial equipment is available in only limited capacity.	• Liquid milk processing (with heat) to produce "pasteurized", ESL, long-life milk. • Pretreatment of milk for cheese manufacture. • Pasteurization of products containing bioactive components.
Ultrasonication (Noci, 2017; Deeth et al., 2013)	• Less energy consumption during product shelf life enhancement by microbial inactivation without inducing much changes in physicochemical and nutritional properties. • Reduces homogenization time and temperature. • Decreases dissolution time of the process. • Controls dairy viscosity and prevents age thickening. • It accelerates fermentation, decreases undesirable flavor and results in better quality products. • At low pressures (more cost effective), it improves flux during microfiltration and ultrafiltration. • Noninvasive and nondestructive, fast and inline diagnosis without product contact • Shelf life enhancement of dairy products.	• Deposition of metal particles in product from sonotrode. • Undesirable chemical changes or degradation of some nutrients (ascorbic acid), lipid oxidation and development of off-flavors. • Ultrasound alone can only inactivate vegetative cells, but not spores and enzymes, hence for better results, it must be used in combination with temperature and pressure.	• Microbial inactivation. • Homogenization. • Cutting and dissolution. • Viscosity reduction. • Fermentation, filtration aid/fouling reduction. • Extraction. • Monitoring. • Degassing/foam reduction. • Reducing heat stability of spores.

Applications of Ultrasonics in the Dairy Industry

Ultrasound applications are governed by three methods, namely direct application to the product, coupled with the device and submersion in an ultrasonic bath. According to Deeth et al. (2013), applications of ultrasound in the dairy industry include filtration, defoaming, fermentation, alteration in viscosity, inactivation of microorganisms and enzymes, crystallization, pasteurization and sterilization, emulsification, homogenization etc. The combined use of ultrasound and pressure, ultrasound and heat, ultrasound, pressure, and heat are known as manosonication, thermosonication and manothermosonication, respectively.

Ultrasound alone can only inactivate vegetative cells, but when combined with heat can inactivate vegetative cells and spores. Moreover, combined use of ultrasound, heat and pressure can inactivate vegetative cells, spores, and enzymes. Ultrasound alone is not suitable for milk pasteurization and leads to cooked/off-flavor development in milk (Mason et al., 2005). Ultrasound also induces homogenization of milk fat, with the formation of stable emulsions, and can improve crystallization of milk fat.

Ultrasonication results in better fermentation of milk with yoghurt culture; it accelerates the rate of hydrolysis of lactose and decreases fermentation time, and also improves the water-holding capacity and firmness of the final yogurt (Wu et al., 2000). Ultrasonication also decreases fouling on membranes, metal surfaces of heat exchangers and extruders and enhances the efficiency of spray driers (Deeth et al., 2013). So far, the industry has adopted only a few applications of ultrasound for commercial use.

Conclusions and Perspectives for the Dairy Industry

Table 3 shows the advantages, disadvantages and possible applications of selected non-thermal technologies in the dairy industry. The different processes described here are at different stages of maturity and adoption by the dairy industry. HPP products such as fruit juices are becoming increasingly common for reasons of minimal processing combined with health and safety aspects; however, the production of HP-treated milk and milk products on an industrial-scale is limited. High-pressure homogenization has shown potential for use in the dairy industry, but so far has found only a few applications in dairy processing, probably due to the availability of only small capacity equipment. PEF is not only able to pasteurize milk in a continuous manner but also allows for retention of important nutrients and the sensory attributes of fresh milk. For commercial pasteurization of milk, PEF and thermal treatment have been suggested as effective and energy-efficient processes. Ultrasound offers many advantages in dairy processing, such as microbial reduction and tailoring functionality of dairy ingredients, but still, its uptake in the dairy industry is not widespread. So far, these non-thermal processes in isolation are suitable for some specific applications; however, their use in combination with other thermal and non-thermal processes can improve the overall efficacy and expand the applications base.

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