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Heat Induced Changes in Milk and Traditional Milk Products

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Authors' contributions

This work was carried out in collaboration between both authors. Author DM wrote the first draft of the manuscript and managed the literature searches. Authors IB revised the manuscript and analysed the study. Both authors read and approved the final manuscript.

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ABSTRACT

Milk is subjected to various heat treatments from mild to severe conditions, with primary objective to enhance the product shelf life and to ensure the food safety. Study on heat induced changes to milk will have significant knowledge on nutritional aspects of the milk and some dairy processing techniques can be altered to reduces the negative impact of heating on milk and milk products. Several unidentifiable milk residues were found while processing and their effect on milk digestibility was not yet studied in detail. As infant milk formula is constituted with concentrated of milk protein and milk bioactive substance, insights on changes to these components would be inevitable. Thermally induced changes also greatly affect the rheological and sensory properties of dairy products. Functional properties of protein such as solubility, emulsion formation and foaming capacity are influenced by heating. Heating conditions leaves behind certain reversible and irreversible physio-chemical changes to milk constituents. This chapter emphasis the heat induced changes in milk constituents milk protein, fat, lactose and salts in an elaborate manner; since studies on product chemistry was focused, changes in bioactive compounds were not given importance. Concern was extended to allergenicity, cross-linking and digestibility of milk constituent wherever possible.

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1. INTRODUCTION

Milk is a biological complex of various components that are highly heat stable. In current dairy processing sectors, milk has been subjected to various heat treatments from mild (thermization, pre-heating, Low Temperature Less Time (LTLT) pasteurization) to severe conditions (Ultra-high heat treatments, thermal drying, sterilization) to produces different dairy products. Heating is considered as an essential processing step with primary objectives of destruction of psychrotrophs, vegetative cells, microbial enzymes and all bacteria (in case of sterilization) to manufacture dairy products within the ensured food safety regime. Heating of milk and milk products causes certain changes in fat, protein, salts and other biologically significant components. This chapter briefs the physico-chemical changes of milk and milk products due to heating in detail.

2. HEAT INDUCED CHANGES IN MILK FAT

2.1 Changes in Fat Phase

2.1.1 Creaming

Milk is a least affected component during heating process, however, principal changes in physico-chemical behavior are creaming and formation of free fat. Creaming is accelerated in the presence of cryo-globulins (a group of immunoglobulins) and the phenomenon is called as cold agglutination. These cryo-globulins particularly enhance the creaming process in cow milk and but not effective in buffalo or ovine milk. There are two types of cryo-globulins, one is denatured by heating (70 °C × 15 min) and the other one is denatured by homogenization. Fortunately, both the cryo-globulins are denatured, that further prevents agglutination in heated milk [1].

2.1.2 Inactivation of lipolytic enzyme

Lipase causes lipolytic rancidity to milk and milk products, especially produced by psychotrophs like *Pseudomonas* and *Bacillus* species. Heat treatment of milk (thermization) significantly reduces the activity of *Pseudomonas* and *Bacillus* species and other psychotrophs, but the lipase enzyme produced these microbes are highly heat stable. Whereas, heat stable Lipoprotein Lipase (LPL) enzymes gets denatured or inactivated at HTST pasteurization. But some studies states that residual activity of LPL found at the level of 64% [2] and 50-74% activity in UHT treated milk [3].

2.1.3 Effect on fat oxidation

Heat treatment causes disruption in MFGM and dissolves the Copper from membrane to serum phase, that acts as a pro-oxidant and in-turn increase the oxidative stability of butter manufactured from it. Formation of free fat occurs when milk fat is subjected to severe heat treatments like spray drying and roller drying. Free fats results in oiling off or cream plug defects in milk powders. The expression of free fat increases the fat oxidation, reduces the dispersibility and wettability in milk powders [1].

2.1.4 Isomerization of conjugated isomers

At high heating temperature (200- 220°C), Linoleic fatty acids (cis-9,cis-12 C18:2) undergoes geometrical isomerization to form their conjugates via auto-oxidation of methyl linoleate chemical pathway [4]. Conjugated Linoleic Acid (CLA) is an intermediate product formed in the reduction process (biohydrogenation), during ruminal biosynthesis of CLA. Among dairy products, processed cheese contain high level of thermally induced CLA content (eg. 8,810.7 mg/kg CLA in fat in Cheese whiz) [1]. Modern cooking methods like microwave heating significantly reduced the CLA even at low temperature, consequently microwave heating of milk caused increase in trans fatty acid isomer (31% in raw milk and 19% in pasteurized milk, that would be potentially undesirable and unhealthy on consumption. Also, microwave heating of cheese would be detrimental because the depletion of CLA was 53%, where the product loses its health beneficial effects [5].

2.2 Changes in MFGM

2.2.1 Interaction with whey protein

As whey protein is a heat liable protein, it goes to denaturation phase at different temperature, unfolding protein starts from 52°C for Bovine Serum Albumin (BSA) followed by aggregation at 60°C, α- lactalbumin (α-la) and β-lactoglobulin (β-lg) undergoes changes above 100°C/10 min, this phenomenon is highly pH- dependent [6]. β-lg and α-la on denaturation, forms disulphide bond interaction with MFGM when heated above 60°C/10 min, and the level of associated proteins on MFGM were 1.0 and 0.2mg/g of fat in heated whole milk respectively, in two-dimensional SDS-PAGE technique [7].

2.2.2 Compositional changes to native MFGM

Due to de-emulsification process of fat globule through homogenization, the adsorption of skim milk protein (β-lg, to lesser extent α-la and β-casein) occurs by partial replacement of membrane layer. This adsorption mechanism makes β-lg and β-casein resistant to gastric digestion [8]. While the native components like XO (Xanthine oxidase) and BTN (Butyrophilin) was unaffected, and found to be intact at both pasteurization and UHT treatment at the temperature of (85°C) and UHT (125°C -145°C) [9]. While heat liable minor components like PAS 6 and 7 where desorbed from the surface of MFGM, due to the reason of attachment of β-lg to hydrophobically exposed bonds of polar lipids. Proteomic study by [10] on heat treated MFGM revealed, that un-identified protein fractions (BTN1A1, ALDH2 and Peroxiredoxin-5) were increased proportional to intensity of heat treatment and almost 50% of the MFGM protein were not able to identify after spray drying [11]. The biological role of these fractions in human health has not been studied in detail. Also, native proteins LBP, cathepsin D, and lysosomal alpha-mannosidase was found intact [12].

3. HEAT INDUCED CHANGES IN PROTEIN

3.1 Changes in Casein

Generally, Casein (CN) is a heat stable protein when treated in its natural state. But thermally induced changes is highly pH dependent.

3.1.1 Hydrolysis of casein

Casein can be hydrolyzed completely into amino acids in the presence of acidic agents (6M HCl) at110°C. Heat treatment in neutral pH causes casein to undergoes lesser level of hydrolysis, formation of peptide and free amino acids were occurred. Casein when heated above 135°C/60 min [13], liberated soluble nitrogen especially at higher rate from α-CN than β-CN in whole milk at normal conditions. And this liberation of N was due to heat treatment that alters the neutral condition to acidic, where NH_3 (ammonia) being formed during hydrolysis, resulted in higher NPN (Non Protein Nitrogen) content. Consequently, the higher NPN was also due to release of peptides (low molecular weight ~2600 Da) having tyrosine and/or tryptophan amino acids [14] and certain peptides were found similar to casino-macro-peptides (CMP) from k-CN (50-100°C). Higher deamidation levels were observed at higher reaction temperatures (90–140 °C) and with increased holding times (up to 30 min) at each temperature. Increasing the phosphate concentration and increasing the pH from 6.0 to 10.0 also increased the level of deamidation; however, increasing the calcium chloride level from 2.5 to 7.5 mM decreased the level of deamidation whereas sodium chloride had no effect even at much higher concentrations/ionic strengths (30–90 mM) [13]. On hydrolysis study on milk and casein micelles, α_{S1}-CN was highly vulnerable to thermal induced hydrolysis than β-CN and k-CN.

3.1.2 Heat induced deamidation

On heating above 90°C with minimum holding period of 30min, amide group like Asn (Asparagine) and Gln (Glutamine) engage in deamidation through acid/base catalysed hydrolysis reaction. The process depends on processing temperature, time, pH and mineral content of milk and milk products [14]. This deamination products acts as a one of the factor in influencing the NPN content of the product [15]. Deamidation of protein and peptide molecules has been proposed to have biological role in human degenerative diseases and ageing process [16]. But the beneficial significance of deamination of milk protein either by enzyme-catalysed or thermally induced process have not been studied yet. Further research on deamidation of milk protein and peptides could widen the use of deamide milk protein as a nutraceutical ingredient.

3.1.3 Heat induced dephosphorylation

Complete dephosphorylation of α and β-CN can be achieved when heated above 140°C/1 hr and no reaction occur till 91°C/1 hr. The rate of dephosphorylation is directly proportional to increase in temperature and moderately affected by pH. Generally, the effect of phosphorylation was observed to be more severe on α-CN than β-CN. Milk processing techniques like preheating has no influence on dephosphorylation and double the rate of dephosphorylation was observed during milk concentration. Enzymatic dephosphorylation of casein showed increased the digestive capacity in infants and improved acid clotting rate [17]. Dephosphorylated whole bovine casein (modified casein) had reduced forming property and increased solubility in their isoelectric point, it also had great emulsifying action at the concentration of 40% modified casein, and has several functional properties in food industry [18].

3.1.4 Formation of Dehydroalanine (DHA) and its interaction with amino acids

DHA is responsible for the wide range of protein modification. DHA was formed during heating of whole casein (especially β-CN) at 80-140°C with pH of 6-10 at various holding period. The concentration of DHA increased with increase in temperature, and found to be higher in phosphorylated β-casein than dephosphorylated β-casein (10-20% reduction). β-elimination occurs in amino acids like serine, cysteine and cystine and residues of serine (serine phosphate and serine gylcosylate) to form DHA [19]. As DHA is highly reactive and unstable molecules it reacts with several other amino acids like lysine, histidine cysteine and theronine to form lysinoalanine, histidinoalanine, lanthionine and methyl-dehydroalanine (further reaction with lysine and histidine to form lysinomethylalanine and histidinomethylalanine respectively). But, the primary source of DHA formation was due to hydrolysis of phosphoserine residues than β-elimination of amino acids [20].

3.1.5 Formation of Isopeptides

Isopeptides does not occur under normal thermal conditions in dairy processing, only formed at severe heat treatments (120°C/24 h) in the presence of more than 4 proteinase and peptidase. Complete hydrolysis of α-amide bond from asparagine and glutamine amino acids and their acid results in aspartyl-lysine and glutamyl-lysine respectively [21]. The concentration of aspartyl-lysine was found to be 4-fold higher than glutamyl-lysinewhen treated at 115°C /27 hr. These isopeptides interfere with protein via cross-linking by forming polymeric chains and thus significantly reducing the protein digestibility [22].

3.2 Changes in Whey Protein

3.2.1 Decrease in solubility

Moderate heating (60-90°C) of whey protein causes structural modifications such as unfolding, where the globular conformation was irreversible and exposure of disulphide bond. Whereas, higher heat treatment causes subjection of hydrophobic bonds that involves in protein cross-linking, thiol oxidation and protein aggregations at extreme [23]. The most potential advantageous measure of changes in whey protein or denaturation rate of whey protein is solubility. Upon heat treatment, the solubility decreases in the order of α -la > β-lg> BSA> Ig. However, there was an unsettlement in this order when observed under DSC microscopy, Ig > β-lg > α -la > BSA. This confusion was due to the renaturing capability of α -la, for detailed information refer [1].

3.2.2 Heat induced aggregation

Aggregation of protein is a physical phenomenon, whey protein undergoes 3 different type of aggregation such as Type I aggregation - characterized by the occurrence of intermolecular disulphide bonds, Type II aggregation - driven by non-specific interactions that include hydrophobic and electrostatic bonding and Type III aggregation- Non-specific interactions with hindered sulphydryl entities.β-lg speed ups the aggregation process of α -la via intermolecular thiol and disulphide interactions and forms as catalyst for heat aggregation [24]. The size and number of voids in whey protein aggregates are significant in the degradation of enzymatically stimulated proteins [25].

3.2.3 Heat induced carbonylation

Carbonylation is the direct measure of oxidative deterioration to the protein. Amino acids like His, Lys, Met, Gln, Pro, Val, Cys, Tyr, Trp and Asn are highly prone to oxidation. Oxidative studies on whey protein isolate showed that, ditryosine, a highly reactive substance formed during oxidation of tyrosine thereby tyrosyl radical as intermediate compound, that continues oxidation on further storage [26]. As other components, heating period and temperature determine the level of oxidation (production of carbonyls). Upon different thermal processing methods, microwave heating of WPI increased the tyrosine and carbonyl formation when compared to freeze drying and conventional drying methods [27]. Oxidation of whey protein has negative impact on the digestibility and bioavailability of the whey protein due to formation of amide bond [28].

3.2.4 Effect of heat on whey protein digestibility

Whey protein in its native globular conformation is highly resistant to gastric digestion; whereas heat treated protein was digestible to acidic conditions of stomach, due to exposure of cleavage and unfolding that enables gastric enzymes to act. Whey protein components, α-la and β-lg showed similar digestive pattern, but pasteurization (above 75°C) and homogenization negatively impacts digestion of β-lg. Heated β-lg has proven to increase the body mass of the rats comparatively to the native β-lg [29]. Degree of glycation, cross-linkage, whey protein aggregation RR induced by maillard reaction increased its digestive capability; aggregation was highly dependent upon pH and heating time [11]. Hence, conscious on heating period, pH, structural deformation, porosity of whey protein RR and its products has to be given greater importance to deliver the desired nutritional and immunological benefits of through final product. On dry heating WPI form conjugates with lactose with the aid of maillard reaction [30], thus improving the emulsifying property to form stable oil-in-water emulsions. Whey protein hydrosylates also form conjugates with maltodextrin, gum acacia, pectin and dextrin at various degree of polymerization.

3.2.5 Heat induced cross-linking

occurs in pH modified environment either through maillard reaction or Dehydroalanine (DHA) process. The primary amino acids involved are Lys, Cys and His in protein to form cross linked protein fractions like lysinoalanine (LAL), lanthionine (LAN) and histidinoalanine as a result of β- elimination, respectively, which are stable during storage of product. These degradation products increase with increase in heating temperature as evident in WPI. Various bonds are responsible for cross-linking S-S bond (disulphide bonds), H bond and other covalent bonds paired with free sulphydryl residues. β- lg cross-linking occurs in raw milk when heated at the temperature of 90°C /30 min due to S-S bond, a type of disulphide interaction; whereas, non-disulphide bonds was reported in maillard reaction when heated above 120°C and formation of isopeptides [31]. Cross-linking of protein causes trypsin resistant digestion and reduced bioavailability, due to the fact that certain part of protein cannot be digested by the gastric enzymes [32] by conformational changes, this event can be positively used in industrial application for development of digestion resistant nutraceutical delivery or slow releasing of desired product encapsulated with heat treated protein.

3.2.6 Heat reduced allergenicity of whey protein

Cow millk allergy is the major concern of the dairy industry mainly caused by a heat sensitive protein β-lg and to lesser extent by α -la and lactoferrin [33]. Pasteurization temperature of 65°C/30min denatures the β-lg allergenic epitopes due to CN and whey protein aggregation and completely losses its activity at 75°C/30 min. Allergenic reaction of β-lg was affected by the presence of lactose [34]. Spray drying processing of milk powder combined with hydrolyzed lactose reduced IgE binding capacity of the protein in milk and formation of glycated products further reduced the allergic conditions [35]. Glycation of whey protein when supplemented with reducing sugars siginificantly reduced the allergenicity. As glycation results in production of AGEs which was confirmed to be harmful, heating without the step of glycation process would be beneficial. As a result of this process, pretreatments like ultrasonication at 400W and high pressure microfluidization [36] decreased the allergenicity of β-lg and α –la by reducing the IgE binding capacity without the formation of glycated products; unfortunately this process said to increase the number of glycation susceptible sites [37] . However, reduction of allergenicity of the whey protein through various pretreatments were not satisfiable, and yet further research should be done to reduce the producing of glycated products and glycation sites.

4. HEAT INDUCED CHANGES IN MILK SALTS

Studies on heat induced effect on milk salts equilibrium are extremely tedious, since salts occur in both aqueous and colloidal phase; on heating salts shift from colloidal phase (complexed with casein micelles) to aqueous (as free ions or polyvalent ions) and some salts renatures to colloidal state on cooling. Salts associated with casein are called micellar or colloidal or micellar calcium phosphate (MCP that includes Serine-phosphate and casein-serine) and the aqueous salts as Donnan ions. Only the aspects of ultrafiltration, microfiltration of whey and milk systems were discussed below, as these 3 processing steps are majorly used in dairy industry when milk salts play an important role.

4.1 Changes during Ultrafiltration

Ultrafiltration is a major dairy process employed in cheese making, production of whey protein concentrates and caseinates etc. During the process, milk is separated into salts and lactose solution. Milk permeates at 25°C started to show the signs of calcium phosphates precipitation and further increase in temperature of 37°C lead to crystallization of Ca and P. Heating above 90°C/30 min caused 60% of Ca and 30% of P was completely precipitated as fine amorphous salts crystals, this heat precipitation should also be consider along with maturation rate of salt to reach its thermodynamically state [38]. Complete removal of MCP from the system is impossible through ultrafiltration or dialysis due to the complex nature of P as it was presented as organic, inorganic and esterified complex with CN and phospholipids.

4.2 Changes during Microfiltration

Microfiltration of whey that contains most of the Ca and P on heat concentration, reaches the saturation rate followed by precipitation. Conversely, the precipitation of CaP was reported to be strongly inhibited by β-CN, protease peptone (PP) 5 and PP8 fast. The rate of turbidity from MF milk permeates was less than compared to UF permeates, due to less Ca and P and precipitation do proceeds [38]. This stabilization of amorphous salt was due to the formation of phosphate centers present in CN micelles as calcium phosphate nanoclusters. Stabilization was also enhanced by presence of Mg, citrate, β-CN, PP5 and PP8 fast.

4.3 Changes in Milk System

Raw unheated milk at 38°C (as in udder) showed the presence of CaP nanoclusters with the radius of 2.5 nm, encored by CN-serine residues as a form of complete protective shell to prevent from crystallization [39] . Warming of milk to 40°C, majorly affected the serum minerals, whereas the colloidal state remained least affected. One set of study reported by [40] states that heating cow milk to 40°C and 80°C resulted in loss of 10% and 80% of colloidal calcium respectively. Exchange of salts between serum and colloidal phase occurs at 90°C, especially P_i (inorganic phosphate) exchange within short period of heating. When heated above 90°C, this exchange completely becomes irreversible. In concentrated milk, the ratio of Ca/ P_i 1.5 gets higher due to the possible reason of amorphous state to crystalline state of the salts [41].

5. HEAT INDUCED CHANGES IN LACTOSE

5.1 Heat Induced Maillard Reaction

Non-enzymatic glycation process of lactose, involves in reaction between the carbonyl and Lys groups of amino acids to form Lactulosyl-Lys complex [42] , thereby resulting in Amadori rearrangement, that produces several compounds which is subsequently hydrogenated and reduced to form dark brown melanoidins. This reaction was desirable in baking, smoking and production of milk based sweets, but not acceptable in milk and concentrated products like milk powder, evaporated milk, soups and pasta products. The undesirable effects in milk system was due to the formation of 5-HMF (Hydroxy-Methyl-Furfural) which was reported to be cytotoxic, hepatotoxic, nephrotoxic, mutagenic, oncogenic, and allergen in nature [43] . Thermal study on degree of glycation in lactose related to heating was presented by [11] , stated that degradation indicators like furosine (formed by acid hydrolysis of Lactulosyl-Lys in heated milk) and 5-HMF was steadily raised when heated above 105°C and reached its maximum at 120°C when heated for 30s. (furosine 255.3 ± 19.7 mg/100 g of proteins and 5-HMF 1.65 ± 0.119 mg/kg when compared to raw milk 3.9 ± 0.7 mg/100 g of proteins and 0.12 ± 0.022 mg/kg respectively). Hence, heating milk above 120°C for 30s should be avoid as much as possible to avoid the detrimental effect of harmful substance formed due to maillard reaction of milk.

5.2 Heat Induced Formation of Lactulose (LCT)

An isomer and epimer of lactose, quantified only in severe heat heated milk like indirect sterilized milk and UHT milk, while completely absent in raw milk. Only 20% lactose undergoes maillard reaction, while 80% experience Lobry de Bruyn-Alberda van Ekenstein process during thermal processing [44] . LCT manifested the conformational structure on Arg and Lys residues of β-lg by reducing its mobility. Due to the alteration of trysin sites and cross linking of protein, digestive capability of the protein has imapaired. LCT can be quantified using HPLC and capillary zone electrophoresis [45] , various studies [11] reported that the concentration of LCT increased when heated above 105°C (16.2 ± 0.7 mg/L) and reached maximum at 120°C for 30s (1057.65 mg/L). Eventhough LCT was considered as a prebiotic and bifidus factor, advanced glycation end products (AGEs) formed at higher temperature affects the digestibility, solubility and emulsifying capacity of casein and whey protein by conjugation [46].

5.3 Heat Induced Advanced Glycation End Products (AGEs)

AGEs occurs in processed foods through maillard reaction. These products became an emerging threat to food industry, as they results in potential human risks like CVD, neuro-degenerative conditions, diabetes and kidney failure. AGEs consist of both low and high molecular weight compounds such as Nε-carboxymethyl-lysine (CML) and Nε-carboxyethyl-lysine (CEL), Pentosidine, Pyrraline [47] and dicarbonyl products like glyoxal and methyl glyoxal and their secondary compounds [48] (Eggen and Glomb 2021). On storage, especially in infant formula, the concentration of CML increases when the package was stored in higher humidity and adverse environmental conditions [49] . 14 types of AGEs products are formed even in the severe heat treated raw milk [50] and soy based infant milk formula had larger concentration of Arg residues that resulted in higher dose of methylglyoxal hydroimidazolones (MG-H) and glyoxal hydroimidazolones (G-H) when compared to cow, and goat milk formula [51]. Hence, the consumers of infant formula should be directed by the manufactured to store and heat the product with high precautions to prevent the glycation reactions.

5.4 Heat Induced Formation of Organic Acids

Heating of milk above 100 °C, resulted in production of early maillard reaction products that are reversible in nature. Galactose and formic acid are the principle compounds [52], while the minor products were acetic acid, citric acid, pyruvic acid, formic acid, succinic acid and oxalic acid. Formation of formic acid induced increase in pH change 6.6 to 6.4 when heated for 120°C/20 min. The possible route for formic acid formation was identified due to degradation of various compounds like galactose, lactulose and lactosyl-lysine. Other organic acids were found to be at low concentration. UHT treatment of milk produced lactic acid at the level of 3.45 mg/100g (150 times higher than typical pasteurized milk) relatively higher than other acids formed. The concentration of these acids were increasing steadily on subsequent storage period [53].

6. HEAT INDUCED CHANGES IN TRADITIONAL DAIRY PRODUCTS

6.1 Khoa

Khoa is a heat desiccated Indian dairy produyct, also called as mava or kawa, made from mixed milk or cow or buffalo milk with continuous stirring and scraping under high heat, until grainy texture, typical light yellow color and caramel flavour is obtained. Khoa is also used as a base ingredient for manufacture of other products like gulabjamun, kalakand, milk cake, burfi and peda, where it is subjected to further processing steps. Desiccation was the primary goal for making of khoa, removal of water causes dehydration of protein, heat denaturation, protein coagulation and exposure of hydrophobic bonds and disulphide bonds, surface hydrophobicity to produce cooked flavour [54]. Heating milk to 132°C - 136°C causes CN denaturation and below 100°C causes whey protein denaturation, structural studies shows that CN-whey protein agglomerates precipitates to produce gritty and grainy texture. Restriction in water activity causes lower redox potential [55] that increases the chance of lipid oxidation. Continuous stirring and scraping induces the rupture of MFGM and formation of free fat. Milk protein and lactose forms complex on heat stress, lactulosyl-Lys forms as a indicator of early maillard reaction and browning indicators like HMF, lactulose and furosine were identified [54]. Sandiness in khoa can be prevented by high speed brisking motion at the end of heating process, that results in smaller lactose crystals formation. Changes in milk salts was significantly as it determines the heat stability of milk due to change in salt balance, to counter the adverse condition soluble Ca ions moves to colloidal state as the aqueous phase was minimizing. Typically, all the salts concentrates into colloidal phase increasing the ash content of khoa [56]. Formation of carbonyl was reported by [57] and [58] like methyl ketones, 2,4dienals, and alk-2-enals were steam distillated and methyl ketones was found to be high in concentration. SEM images of khoa revealed that lactose and protein forms complex and network appears with many void, that gives khoa its soft texture [59].

6.2 Ghee or Clarified Butter

Heat concentrated fat product prepared by creamery butter method industrially. The process involves concentrating milk fat into cream and conversion into butter by continuous butter making machine, followed by heat concentration of butter to ghee or clarified butter. The changes in only fat were left in ghee to be discussed. As MFGM gets ruptured and demulsification occurs while butter making, this unsalted butter will be subjected to different heating temperature, initially heat to remove water content by preventing frothing of milk protein [55], than heated to 103°C therefore denaturing the residual lactose and milk protein (ghee residue), production of AGEs, advanced maillard reactions and flavour compounds like lactones, methyl lactones, carbonyls and fatty acids. The heating temperature should be controlled to prevent the development of off flavour and charring of ghee residues. When finally heated to 108°C, the bounded water vaporizes with residual moisture content of not more than 0.5% [60] , ghee residues charred and forced to suspend at the bottom of clarifying tank (at 110°C). Conjugation of unsaturated fatty acid leads to [61]heat induced isomerization of Linoleic acid to Conjugated Linoleic acid (CLA) with the concentration of 600mg per 100g (90% of cis-9, trans-11 and 10% of trans-10, cis-12 isomers). Granulation of fat crystals is an important quality attribute of commercial ghee, [62] studied that cooling of ghee to 22°C causes nucleation and crystal growth of high melting TGAs (triglycerides).

6.3 Paneer

Paneer is a heat treated and acid coagulated dairy product and also used as a base ingredient for preparation for various Indian savoury cusinies and curries. Paneer (preferably from buffalo milk, as the yield is high compared to cow milk) resembles the textural and micro-structural characteristics of cottage cheese, whey protein gets denaturated when preheated to 90°C (coagulated at 70°C) that enhances the water holding capacity of β-CN, and so increasing the yield of paneer. Solubility of colloidal calcium phosphates will be restricted and formation of acid curd with co-precipitates of β-lg and k-CN occurs. Heat treatment of milk determines the rheological properties and yield of paneer. Ultrastructure of paneer reported by [63] showed that core-lining structure of larger protein molecules with strands of MFGM attached to it along with clusters of compact smaller protein molecules. The heating temperature greatly affected the core-lining structure, well stabilized lining at high temperature (90°C) due to complex of β-lg - k-CN and partial lining at low temperature. Fat globules were resistant to paneer manufacturing retaining its globular structure. While cooking paneer at higher temperature (above 100°C for making curries) the core structure disintegrates and lining structure predominant. Frying of paneer in vegetable oil causes swelling of protein matrix, maillard reaction and increased water activity [64].

6.4 Gulabjamun

Gulabjamun is a deep fried Indian delicacy, made with khoa dough with starch (refined wheat flour) as base ingredient fried in ghee or vegetable and soaked in 60% sugar syrup with added flavour. Khoa when fried in ghee showed several heat induced manifestation in structural aspects. In continuous deep frying of khoa dough at 136-140°C, water vapour transmission rate increases, volume expansion due to water vapour that form inner porous structure of dough. Two significant changes occurs in deep frying protein coagulation and pregelatinization of starch [65] . The SEM analysis of defatted gulabjamun was evident that fuzzy agglomeration formation of protein and starch suspended as thread like structure loosely bounded around the protein molecules. Fat molecule gets embedded between fused protein and pregelatinizied starch, and retains it globular structure. Starch and fat acts as a weak plasticizers that stabilizes the structure of the product by filling the large void spaces and densely packing the protein with starch [66] to pronounce the soft chewy texture on soaking. The outer surface of the dough (crust) turns golden brown colour – a non-enzymatic browning mechanism that depends upon the intensity of frying. Caramelization of lactose was also one of the factors responsible for crust browning.

7. HEAT TREATMENT AND THE IMPACT ON NUTRITIONAL VALUE OF MILK AND MILK PRODUCTS

The fat, fat-soluble vitamins, carbohydrates and mineral' of milk are essentially unaffected by heal treatment. Proteins and water-soluble vitamins are the components which are mainly affected by the heat treatments. The impact of heat treatment in yogurt production on acidification rate and incubation time is widely recognized. Consequently, the decrease in incubation time can be attributed to the denaturation of whey proteins. The more significant the denaturation of whey proteins, the quicker the incubation period is achieved. However, heat treatment remains the primary factor that determines the span of the shelf-life. Pasteurization and ultrahigh temperature processes altered the composition of the milk slightly, decreasing total fat and total solids and increasing urea [67].

8. CONCLUSION

Heat induced changes in milk and milk products would be significant in designing and optimizing the process parameters for manufacturing the dairy products industrially in larger units. Especially Indian traditional dairy sweet, involves complex and multiple processing procedure that requires manpower inevitably. The changes include both advantageous and disadvantageous alteration in milk constituents. Example formation of AGEs in infant milk formulations and dairy products were extensively given importance in recent studies that negatively impacts the human health. Research on further changes in molecular level would be beneficial to predict the changes and develop the methods or procedure of manufacturing step to prevent that from happening. Since, heating is the primary step for all food products, complex formation of milk protein and other constituents with non-dairy ingredients shall be studied as the digestion and bioavailability of milk protein are greatly affected. The presented article has given the concern on allergenicity, complex formation of milk constituents with recent research from past decade. Knowledge on heat induced changes in traditional dairy products would be helpful to further study on unidentifiable products formed during processing and develop appropriate optimization of process and parameters.

HIGHLIGHTS

1. The review article details the changes in milk components during heating process at molecular level. Study on heat induced changes would be useful for manufacturers of milk powder and Indian sweets to reduce the formation of harmful substance that produces during heating at very high temperature.
2. The allergenicity of certain proteins and effect of heating to reduce the allergen has been discussed, especially in infant formulation.
3. Advanced Glycation End products (AGEs) a new emerging concern in food industry, that is formed due to high heating process has been focused in the article elaborately.
4. Heat induced changes in Indian traditional dairy products like Khoa, Ghee or Clarified Butter, Gulab Jamun and paneer were reviewed at the molecular level.
5. So far, heat-induced changes in Indian traditional dairy products have not been detailed in a single access, the authors acknowledge that, this review is the first kind of detailing heat-induced changes in traditional dairy products.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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