

Effect of Ultrasound Treatment on Milk Homogenisation and Particle Size Distribution of Fat

M. Fatih ERTUGAY, Mustafa ŞENGÜL*, Memnune ŞENGÜL
Atatürk University, Faculty of Agriculture, Department of Food Engineering, 25240, Erzurum - TURKEY

Received: 04.04.2002

Abstract: The effect of ultrasound treatment on milk homogenisation was studied. A microscope, equipped with a camera and ocular micrometer, was used to determine the diameters of fat globules and to evaluate the homogenisation efficiency of milk samples homogenised by using ultrasonic and conventional homogeniser. It was found that ultrasound treatment with high amplitude (power) had an important effect on milk homogenisation compared with conventional homogenisation. As exposure times and power levels increased, homogenisation efficiency also rose. The highest homogenisation efficiency and the smallest fat globule diameter were 3.22 and 0.725 μm at power level 100 (450 W) for 10 min, respectively. The fat globule diameters at a power level of 40 (180 W) for 10 min were similar to those of conventional homogenisation.

Key Words: Ultrasound, homogenisation, fat globule

Sütün Homojenizasyonu ve Yağın Parçacık Boyut Dağılımı Üzerine Ultrason İşleminin Etkisi

Özet: Araştırmada, süt homojenizasyonu üzerine ultrason işleminin etkisi araştırılmıştır. Ultrasonik ve klasik homojenizator kullanılarak homojenize edilen süt örneklerinin homojenizasyon etkinliğini değerlendirmek ve yağ partiküllerinin çaplarını belirlemek için kamera ve oküler mikrometre ile donatılmış bir mikroskop kullanılmıştır. Klasik homojenizasyon ile karşılaştırıldığında, yüksek güçte ultrason uygulamasının, süt homojenizasyonu üzerine önemli bir etkiye sahip olduğu bulunmuştur. İşlem zamanı ve güç seviyesi arttıkça homojenizasyon etkinliği de artmıştır. En yüksek homojenizasyon etkinliği (3,22) ve en küçük yağ partikül çapı (0,725 μm) 10 dakikalık işlem zamanı ve % 100 (450 W) güç seviyesinde elde edilmiştir. % 40 (180 W) güç seviyesi ve 10 dakikalık işlem zamanında elde edilen yağ partikül çapı, klasik homojenizasyonda elde edilen değere benzer bulunmuştur.

Anahtar Sözcükler: Ultrason, homojenizasyon, yağ globülü

Introduction

A number of non-traditional preservation techniques are being developed to satisfy consumer demand with regard to nutritional and sensory aspects, convenience, absence of synthetic additives, low energy demand and environmental safety (1). In addition, consumer demand for foods that are fresher, more natural and healthier and that at the same time provide a high degree of safety have increased interest in non-thermal preservation techniques for inactivating microorganisms and enzymes in foods (2-5). Among these, ultrasound has an important role in food engineering due to consumer interest in

minimally processed foods. It is evident that ultrasound technology has a wide range of current and future applications in the food industry.

Ultrasound is of great interest in food processing for many reasons (6). This technique can be generally used in systems which are liquid and can be defined as waves with a higher frequency (7). Floros and Liang (8) noted the use of low intensity high frequency ultrasound for the improvement of food product/processes monitoring due to the acceleration of diffusion. These industrial applications include the texture, viscosity and concentration determination of eggs, meats, fruits and

*Correspondence to: E-mail: msengul@atauni.edu.tr.

vegetables, and dairy and other products; and thickness, flow level and temperature measurements for the monitoring and control of several processes.

The lethal effect of ultrasound has been attributed to the cavitation phenomenon. Ultrasound cavitation is the formation of bubbles or cavities in liquids. The collapse of the bubbles results in intense shock waves that can cause considerable damage to surrounding material (9). When ultrasound is used in combination with conventional heating, the effect of ultrasound treatment increases.

The use of ultrasound in food technology has been a subject of research for many years and there have recently been increasing numbers of studies regarding this technique (9-31). The objective of our study was to investigate the effects of different ultrasound treatments on milk homogenisation and to compare these with conventional homogenisation.

Materials and Methods

Materials

Cow milk was obtained from the dairy farm at Atatürk University, Erzurum, Turkey. The milk was pasteurised to kill vegetative bacteria and inactivate most enzymes by using a plate pasteuriser at 90 °C for 5 min before the homogenisation treatment.

Methods

Homogenisation by conventional homogeniser

An Alfa-Laval homogeniser (Model SH 20) was used to homogenise the milk under a high pressure of 200 bar at 55 °C.

Ultrasonic Homogenisation

An ultrasonic generator (Cole-Parmer Instrument Company, USA), equipped with a 19 mm diameter tip and working at a constant frequency (20 kHz), was used to homogenise 250 ml of milk for each treatment. This generator converts 50 Hz line voltage to a high frequency electrical power of 20 kHz. The ultrasound probe was immersed into the milk at a depth of approximately 3 cm. The experiment was carried out using different power levels. Exposure times were 5 and 10 min for each trial. During ultrasound treatment, the samples were kept at 55 °C by using a water bath. The power level of 100

corresponded to approximately 450 W, and power levels of 20, 40 and 80 corresponded to approximately 90, 180 and 360 W, respectively.

Microscopy

All samples were cooled to 20 °C and analysed under the microscope. Before analysis, the samples were gently shaken to prevent changes in fat globule size.

A microscope with an oil immersion lens (magnification x 1000, Olympus, EH-2) was used. Samples of 10 µl were placed on the slide and the microscope equipped with a camera was used to take photographs of them. The size of the fat globules was evaluated by using an ocular micrometer in 1 µm increments.

Homogenisation Efficiency

The homogenised milk was placed into a graduated cylinder of 250 ml and kept in a refrigerator for 48 h. Then, the fat contents of samples from the upper part, i.e. 1/10 (a), and from the bottom, i.e. 9/10 (b), of the graduated cylinder were determined by using the Gerber method (32). The following equation was used to calculate the homogenisation efficiency (H_E) of the samples:

$$H_E = \frac{a - b}{a} \times 100$$

Statistical analysis

In the study, 4 different ultrasound amplitude levels (20, 40, 80 and 100%) and 2 different exposure times (5 and 10 min) were selected as experimental factors. The analysis was carried out according to a completely randomised blocks design with 4 replications.

Results

A comparison of fat globule diameters obtained by using conventional and ultrasonic homogeniser is given in the Table. As shown in the Table, milk homogenised by the conventional homogeniser had smaller fat globules than non-homogenised milk, and milk homogenised by ultrasound (higher power levels) also had smaller fat globules than milk homogenised by a conventional homogeniser. However, the higher the ultrasound amplitude levels, the smaller the fat globule diameter.

Table. A comparison of fat globule diameters obtained in conventional and ultrasonic homogenisation.

Treatment	Mean	Standard error	Minimum	Maximum
Non-homogenised	5.500a	0.6455	4.00	7.00
Conventional homogenisation	2.625cd	0.2394	2.00	3.00
Ultrasonic homogenisation				
20:5	3.875b	0.4270	3.00	5.00
20:10	3.500bc	0.6455	2.00	5.00
40:5	3.375bc	0.3750	2.50	4.00
40:10	2.375cd	0.2394	2.00	3.00
80:5	2.125d	0.1250	2.00	2.50
80:10	1.250ef	0.1443	1.00	1.50
100:5	1.125ef	0.1250	1.00	1.50
100:10	0.725f	0.8.539E-02	0.50	0.90
Total	2.648	0.2439	0.50	7.00

The averages marked with the same letters are not different from each other statistically ($P < 0.01$). 20, 40, 80 and 100: power levels (%), 5 and 10: exposure times (min).

The best homogenisation and the smallest fat globule diameter (0.725 μm) were obtained at a power level of 100 for 10 min.

The fat globule diameters of milk homogenised by using conventional and ultrasonic homogeniser ranged between 2.0-3.0 and 0.5-5.0 μm , respectively, while the fat globule diameters of non-homogenised milk ranged between 4.0 and 7.0 μm . The fat globule diameters obtained at a power level of 40 for 10 min (2.375 μm) were similar to those from (2.625 μm) conventional homogenisation (Table). Consequently, the fat globule diameter obtained in conventional homogenisation could be achieved by using ultrasonic homogenisation at power level of 40 for 10 min.

Micrographs of milk samples are shown in Figure 1. As shown in Figures 1a-j, as the ultrasound power levels and exposure times increased, the distribution of fat globules was more stable and smaller fat globule diameters were obtained. It was difficult to observe the fat globules at a power level of 100 for 10 min (Figure 1j) because of breakage into very small fragments. There were numerous fat globules smaller than 1 μm at a power level of 100 for 10 min (Figure 1j). However, the fat globules in non-homogenised milk could be easily seen (Figure 1a).

A significant correlation was found between the diameter of fat globules and homogenisation efficiency ($P < 0.01$). The homogenisation efficiency of milk samples versus amplitude levels is plotted in Figure 2.

As shown in Figure 2, as the amplitude levels increased, the homogenisation efficiency of homogenised milk samples also increased. However, the exposure times had an important effect on the homogenisation of milk.

Discussion

The effect of ultrasound on milk homogenisation has been attributed to the cavitation phenomenon. During ultrasonic treatment, the main active force is mechanical in nature, resulting in the formation and implosion of bubbles in a liquid (cavitation) (33). On the other hand, although many aspects of the ultrasound mechanism remain obscure, it can be related to cavitation (formation and violent collapse of bubbles), heating (specific absorption of acoustic energy), dynamic agitation and shear stresses, and turbulence (8).

Ultrasound treatment has a very good homogenisation effect at higher power levels compared to conventional homogenisation. Therefore, ultrasonic

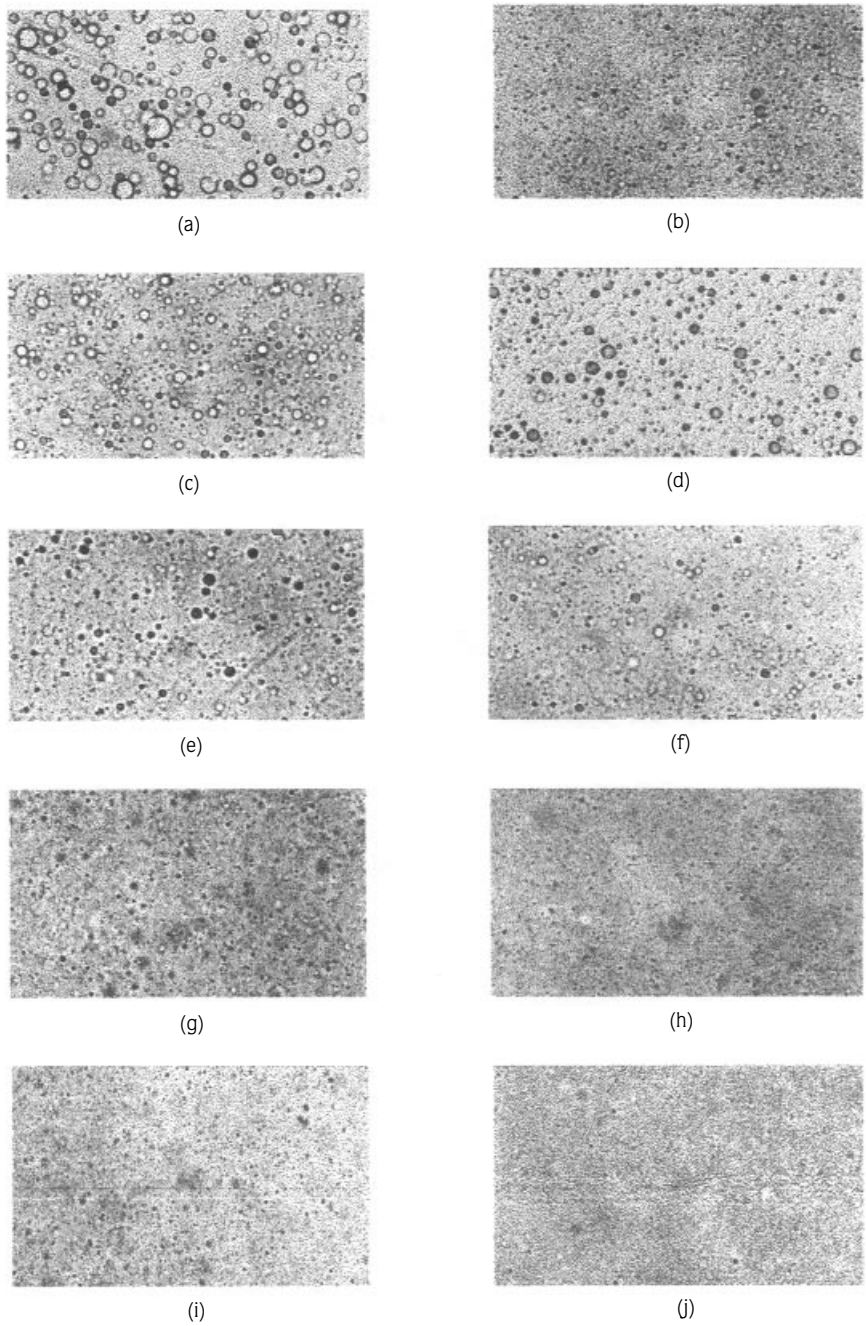


Figure 1. Micrographs of milk samples (magnification x 1000; scale 1 mm). (a) Without homogenisation. (b) Conventional homogenisation under 200 bar at 55 °C. (c) Ultrasonic homogenisation at power level 20 (90 W) for 5 min. (d) Ultrasonic homogenisation at power level 20 (90 W) for 10 min. (e) Ultrasonic homogenisation at power level 40 (180 W) for 5 min. (f) Ultrasonic homogenisation at power level 40 (180 W) for 10 min. (g) Ultrasonic homogenisation at power level 80 (360 W) for 5 min. (h) Ultrasonic homogenisation at power level 80 (360 W) for 10 min. (i) Ultrasonic homogenisation at power level 100 (450 W) for 5 min. (j) Ultrasonic homogenisation at power level 100 (450 W) for 10 min

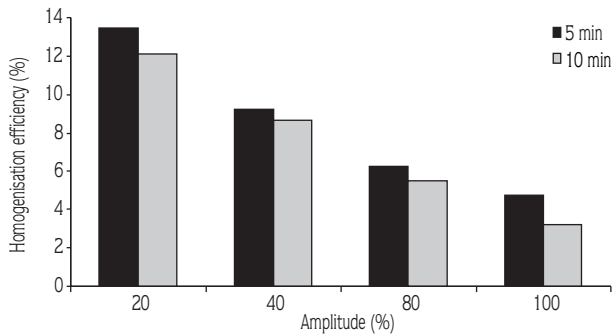


Figure 2. Histogram of homogenisation efficiency vs. amplitude levels.

treatment has been considered as an effective system for the reduction of fat globule size (34,35). However, low power levels had no important effect on milk homogenisation. Homogenisation at a power level of 40 for 10 min was similar to conventional homogenisation. The longer exposure times had a synergic effect on milk

homogenisation. If adequate power levels and exposure times were applied, complete homogenisation could be obtained. Therefore, it can be concluded that ultrasound treatment at high power levels may be used to reduce fat globule size. Schmidt (34) observed that ultrasonic homogenisation of milk at 60 °C could reduce the average fat globule size to 1 µm. Similar results were also obtained in other studies (7,30,36).

Furthermore, the value of homogenisation efficiency must be below 10% for a very efficient homogenisation process (32). A value of homogenisation efficiency below 10% indicates that homogenisation was very efficient. While the lowest homogenisation efficiency was obtained at a power level of 20 for 5 min (13.51), the highest homogenisation efficiency was obtained at a power level of 100 for 10 min (3.22). These results support the results obtained from microscopic analyses.

References

- Alzamora, S.M., Guerro, S.N., Lopez-Malo, A.: Ultrasound as a Food Preservation Method. 2000 IFT Annual Meeting. USA, 2000.
- Mertens, B., Knorr, D.: Development of Nonthermal Processes for Food Preservation. *Food Tech.* 1992; 46: 124-133.
- Gould, G.W.: *New Methods of Food Preservation*. Blackie Academic and Professional, UK. 1995; 159-172.
- Barbosa-Canovas, G.V., Pothakamury, U.H., Palou, E., Swanson, B.G.: *Nonthermal Preservation of Foods*. Marcel Dekker, New York, 1997.
- Manas, P., Pagan, R., Raso, J., Sala, J.F., Condon, S.J.: Inactivation of *Salmonella enteritidis*, *Salmonella typhimurium* and *Salmonella senftenberg* by Ultrasonic Waves under Pressure. *J. Food Protec.* 2000; 63: 451-456.
- Letang, C., Piau, M., Verdier, C., Lefebure, L.: Characterization of Wheat-Flour-Water Doughs: A New Method Using Ultrasound. *Ultrasonics*. 2001; 39: 133-141.
- Villamiel, M., Hamersveld, E.H., Van Jong, P. De.: Effect of Ultrasound Processing on the Quality of Dairy Products. *Milchwissenschaft*. 1999; 54: 69-73.
- Floros, J.D., Liang, H.: Acoustically Assisted Diffusion through Membranes and Biomaterials. *Food Technol.* 1994; 48: 79-84.
- Kim, S.M., Zayas, J.F.: Processing Parameters of Chymosin Extraction by Ultrasound. *J. Food Sci.*, 1989; 54: 700-703.
- Kim, S.M., Zayas, J.F.: Effects of Ultrasound Treatment on the Properties of Chymosin. *J. Food Sci.*, 1991; 56: 926-930.
- Wrigley, D.M., Llorca, N.G.: Decrease of *Salmonella typhimurium* in Skim Milk and Egg by Heat and Ultrasonic Wave Treatment. *J. Food Protec.* 1992; 55: 678-680.
- Samari, S.: Ultrasonic Inspection Methods for Food Products. *Lebensmit. Wissen. Technol.* 1994; 27: 210-213.
- Earnshaw, R.G., Appleyard, J., Hurst, R.M.: Understanding Physical Inactivation Processes: Combined Preservation Opportunities Using Heat, Ultrasound and Pressure. *Int. J. Food Mic.*, 1995; 28: 197-219.
- Benedito, J., Bon, J., Sanjuan, N., Frau, M.: Utilization of Ultrasound in the Food Industry. I. Fundamental Concepts. *Aliment. Equip. Tecnol.* 1995; 14: 79-83.
- Lopez, P., Burgos, J.: Peroxidase Stability and Reactivation after Heat Treatment and Manothermosonication. *J. Food Sci.*, 1995; 60: 451-455.
- Banerjee, R., Chen, H., Wu, J.: Milk Protein-Based Edible Film Mechanical Strength Changes Due to Ultrasound Processes. *J. Food Sci.*, 1996; 61: 824-828.
- Barton, S., Bullock, C., Weir, D.: The Effects of Ultrasound on the Activities of Some Glycosidase Enzymes of Industrial Importance. *Enzyme Microb. Technol.* 1996; 18: 190-194.
- Dahzong, W., Sakakibara, M., Kondoh, N., Suzuki, K.: Ultrasound-Enhanced Lactose Hydrolysis in Milk Fermentation with *Lactobacillus bulgaricus*. *J. Chem. Technol. Biotechnol.* 1996; 65: 86-92.
- Mason, T.J., Paniwnyk, L., Lorimer, J.P.: The Uses of Ultrasound in Food Technology. *Ultrason. Sonochem.* 1996; 3: 253-260.

20. McClements, D.: Ultrasonic Characterization of Foods and Drink: Principles, Methods and Applications. *J. Crit. Rev. Food Sci. Nutr.* 1997; 37: 1-46.
21. Vercet, A., Lopez, P., Burgos, J.: Inactivation of Heat-Resistant Lipase and Protease from *Pseudomonas fluorescens* by Manothermoutrasonication. *J. Dairy Sci.*, 1997; 80: 29-36.
22. Carcel, J.A., Benedito, J., Bon, J., Frau, M.: Application of Ultrasound in the Dairy Products and Derivates Industry. I. Low Intensity Ultrasound. *Aliment. Equip. Tecnol.* 1998; 17: 39-44.
23. Raso, J., Palop, A., Pagan, R., Condon, S.: Inactivation of *Bacillus subtilis* Spores by Combining Ultrasonic Waves and Mild Heat Treatment. *J. App. Microbiol.* 1998; 85: 849-854.
24. Gennaro, L. De., Cavella, S., Romano, R., Masi, P.: The use of ultrasound in Food Technology. I. Inactivation of Peroxidase by Thermosonication. *J. Food Eng.*, 1999; 39: 401-407.
25. Letang, C., Piau, M., Verdier, C.: Characterization of Wheat-Flour-Water Doughs. Part I: Rheometry and Microstructure. *J. Food Eng.*, 1999; 1: 121-132.
26. Mulet, A., Benedito, J., Bon, J., Sanjuan, N.: Low Intensity Ultrasonics in Food Technology. *Food Sci. Technol. In.* 1999; 5: 285-297.
27. Pagan, R., Manas, P., Alvarez, I., Condon, S.: Resistance of *Listeria monocytogenes* to Ultrasonic Waves under Pressure at Sublethal (manosonication) and Lethal (manothermosonication) Temperatures. *Food Microbiol.* 1999; 16: 139-148.
28. Manas, P., Pagan, R., Raso, J.: Predicting Lethal Effect of Ultrasonic Waves under Pressure Treatments on *Listeria monocytogenes* ATTC 15313 by Power Measurements. *J. Food Sci.*, 2000; 65: 663-667.
29. Marcotte, M., Trigui, M., Tatibouet, J., Ramaswamyit, S.: An Ultrasonic Method for Assessing the Residence Time Distribution of Particulate Foods during Ohmic Heating. *J. Food Sci.*, 2000; 65: 1180-1186.
30. Villamiel, M., Jong, P. De.: Influence of High-Intensity Ultrasound and Heat Treatment in Continuous Flow on Fat, Proteins and Native Enzymes of Milk. *J. Agric Food Chem.*, 2000; 48: 472-478.
31. Villamiel, M., De Jong, P.: Inactivation of *Pseudomonas fluorescens* and *Streptococcus thermophilus* in Trypticase Soy Broth and Total Bacteria in Milk by Conventional Heating. *J. Food Eng.*, 2000; 45: 171-179.
32. Metin, M. Süt Teknolojisi. Ege Üniversitesi Basımevi. Mühendislik Fakültesi Yayınları. No: 33. İzmir. 1998.
33. Shukla, T.P.: Microwave Ultrasonics in Food Processing. *Cereal Food World.* 1992; 37: 332-333.
34. Schmidt, H.W.: Untersuchungen zur Ultraschall-Homogenisierung von Milchproben. *Lebensmittelindustrie.* 1995; 32: 173-175.
35. Martinez, F.E., Desai, F.D., Davidson, A.G.E., Nakai, S., Radcliffe, A.J.: Ultrasonic Homogenisation of Expressed Human Milk to Prevent Fat Loss during Tube Feeding. *J. Pediatr. Gastroenterol. Nutr.*, 1987; 6: 593-597.
36. Wu, H., Hulbert, G. J., Mount, J. R.: Effects of Ultrasound on Milk Homogenisation and Fermentation with Yoghurt Starter. *Innovate Food Sci. Emerg. Technol.* 2001; 1: 211-218.