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## Impact of Ultrasonic on some Physiochemical and Technological Properties of Raw Milk

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#### **ABSTRACT**

Fresh buffalo's milk was used in a series of treatments to elucidate the effect of ultrasonication on its physicochemical and technological properties. Samples were divided into four treatments, in addition to the control. Treatments (A and B) were exposed to Ultrasonication with 20 kHz for 5 and 15 min. at 20± 5°C, treatments (C and D) were exposed to thermosonication with 20 kHz for 5 and 15 minutes at 50±5°C.Results revealed a slight increase of fat and protein contents of milk in the treatments A and B, the same trend was observed in the treatments C and D. The content lactose content slightly varied between the raw milk and Ultrasonication treated milk. There are differences in pH values and acidity. The results indicated that ultrasonication and thermosonication treatments increased the rates of synersis and decreased in curd tension of acid curd. Meanwhile, rennet reduced the coagulation time (RCT, sec) and increased wet curd yield by treating with ultrasonication.

Keywords: Ultrasonication, Thermosonication, Raw milk.

#### INTRODUCTION

Today, consumers are in great demand for highquality natural functional products that contain no preservatives and no other chemical additives. Therefore, many non-chemical methods of processing raw materials are being researched to improve production (Barba et al., 2012; Chandrasekaran et al., 2013; Kiprushkina and Baranenko, 2014).

Ultrasound waves defined as longitudinal sound waves of frequency of 20 kHz or more. The low amplitude ultrasonic waves usually used to analyses the material, but it has no significant modify on the material under examination. The high amplitude ultrasonic waves possess the capacity to alter the food material by cavitation so it can be effective in food processing. Currently, ultrasound processing is exploited in the dairy industry. Thermosonication (TS) is a process describing the simultaneous use of low frequency ultrasound waves (20 kHz) along with heat which may exhibit some synergistic effect. In comparison with conventional heat process, thermosonication reduces the temperature needed in the process so the less energy is consumed. Accordingly, Thermosonication (TS) is considered to represent a green and economical technology; this, in turn, makes it a cost-efficient process. In the promising future, Thermosonication can be used as a commercial method to treat and homogenize milk, as it delivers a desired effect in (Ghosh, 2017). The most significant advantages of exploiting ultrasound for food processing, includes: improving the effectiveness of mixing and micro-mixing, faster energy and mass transfer, selective extraction, more rapid response to process extraction control, increased production, and elimination of process steps (Uzma-Altaf et al., 2018).

According to (Mohammadi et al., 2014), proved that the ultra-sonicated milk improves the efficiency of milk, microbial destruction, equipment surface cleaning and also can manage the waste.

Wu et al. (2000) Showed that the practical use of high amplitude ultrasound typically produced a good homogenization effect compared with conventional homogenization. (Juliano et al., 2013; Leong et al., 2014 and Leong et al., 2016).

The high power ultrasound has a significant effect in liquids, as in the mechanical vibration of the medium and moving of the solid particles contained in it, the acoustic streaming can increase the mass transfer in medium (Tho et al., 2007).

Acoustic cavitation in common is a process that use power of ultrasound processing at low frequencies for milk homogenization which consequently increase the viscosity and enhance texture characteristics of fermented products (Sfakianakis and Tzia,2010) So, it considered to be one of the most important processes for the ultrasonic treatment. By changing ultrasonic power and length of sonication, it may allow receiving very fine emulsions and altering size distribution of the fat globules in milk (Zverev and Lobanov, 2005; Ashok Kumar et al., 2010).

The considerable advantage of the ultrasonic homogenisers is promoting their cleaning relative to traditional homogenizers (Ertugay and Sergul, 2004; Ojha et al., 2017 and Leong et al., 2018). Ultrasound treatment also promotes proteins swelling and changes a ratio of free and bound water (Popova, 2013) that is especially important for fermentation process efficacy. Therefore, the aim of this study was to investigate whether ultrasound power and Thermosonication could be used as an alternative processing method for the purposes of adequate microbiological quality assurance and whether they have effect on the properties of yoghurt and cheese mono-factoring.

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#### MATERIALS AND METHODS

Fresh buffalo's milk was obtained from Sakha Animal Production Research Station. Fresh samples (500 ml) were used in an ultrasonic homogenizer processor 4710 series (parmer instrument Co. Chicago, Illionois 60648). The laboratory investigations were carried out at EPCRS Excellence Center and Plant Pathology and Biotechnology Laboratory Department of Agriculture Botony, Faculty of Agric., Kafrelsheikh Univ., Egypt). Samples A and B were treated by ultrasound (US) at the 20 kHz for 5 and 15 min., respectively at the temperature was  $20\pm5^{\circ}$ C. The aerosonication treatments (TS) were applied on ultrasonic processor at 20 kHz for 5 and 15 min in water bath at  $50\pm5^{\circ}$ C (treatments C and D) respectively.

#### Methods:

Fresh buffalo's milk (control) and the examined treatments were analyzed for fat%, protein%, and lactose% by using milko-scan (133 BN Foss Electric, Denmark). The pH value of samples was measured using digital pH meter, whereas the titratable acidity (%) was determined according to AOAC (2005). Specific gravity was detected according to Ling (1963).

## Making Yoghurt and its properties:

Studies on the effects of ultrasonic homogenizer and thermosonication on the acid coagulation properties of milk were carried out. For examining the activity of yoghurt starter 2 ml of yoghurt starter were added to 100 ml of the samples and was incubated at 42°C. The pH values were determined at different intervals of incubation.

Curd tension was estimated according to Chandrasekhara *et al.* (1957) as described by Abd El-Salam *et al.* (1991). The rate of curd synersis was evaluated as described by Mehanna and Mehanna (1989).

## Cheese making properties of milk:

Ultrasonic homogenizer and thermosonication were applied in cheese technology to reveal the effect of ultrasonic treatment of milk on cheese yield, and on the wet curd yield in a model system and on the rennet coagulation time (RCT). (Lopez-Fondio *et al.*, 1996). Total nitrogen content in whey (mg/100 ml) was also detected by Kjeldahl method (Ling, 1963). In triplicate

Statistical analysis was carried and the analysis of variance and Duncan's test as well as average and standard error were carried out using SPSS Computer Program (SPSS, 1999).

#### RESULTS AND DISCUSSION

### Impact of ultrasound on milk components:

The milko-scan was used in this study to determine the probable changes in the composition of both raw milk and of ultrasonication- and thermosonication-milks. The obtained data from milko-scan are summarized in Table (1).

Table 1. Milko-scan results of the different milk composition ultrasound shock of raw milk (Average±SE of 3 replicates).

Enaction Contr	J J	IJS	TS	
Fraction Contr	5 min	15 min	5 min	15 min
Fat% 6.62±0.	11°6.72±0.29	°6.79±0.04°	6.79±0.04	6.74±0.05a
Protein% 4.14±0.0	02°4.19±0.22	<sup>a</sup> 4.19±0.16 <sup>a</sup>	4.19±0.11	4.20±0.19a
Lactose%5.62±0.4	40°5.68±0.21	a5.74±0.08a	5.72±0.09	a5.56±0.13a

Control: fresh buffalos milk

US: samples were treated by Ultrasonication shock 20 kH2 /20 $\pm$ 5°C TS: samples were treated by Thermosonication shock 20 kH2 /55 $\pm$ 5°C Average with different letter due to treatment differed significantly (P<0.05).

Fat content of raw milk show an insignificant increase (P>0.05), compared with control (6.62%) to 5 min and 15 min (6.72 and 6.79%), in order ultrasonic treatment (US) and also from thermosonication treatment (TS) (6.79 and 6.74%, respectively).

Homogenization of fats treated by ultrasonication resulted in decrease in the fat globules (Michelle *et al.*, 2009). Oguz *et al.* (2016) reported that the thermosonication treatment did not influence proximate composition of samples. The potential application of ultrasonication is to enhance creaming of milk fat globules in milk emulsion system, enhance the fractionation of milk fat in dairy system and achieve the separation of larger fat globules from a smaller one (Juliano *et al.*, 2013; Leong *et al.*, 2016; Leong *et al.*, 2014)

The data of raw sample shows an insignificant increase (P>0.05) in the protein content in control (4.14%) to (4.19%) in the US treatment and also from TS treatment (4.19% and 4.20%), respectively. No further significant changes could be rated in protein content of the remainder of the ultrasonic treatment of the ultrasonic treatment (US) and thermosonication treatment (TS) (Table 1).

Chandrapala *et al.* (2012) observed no marked change in casein micelle size, free casein content and soluble calcium concentration. There was a slight increase in soluble whey protein and corresponding decrease in viscosity. Slight decrease in pH "but temporary" of the skim milk sample subjected to ultrasonication treatment for 60 min. (Villamiel and de Jong (2000)

As with the lactose content of milk samples, data illustrated in (Table 1) indicated an insignificant difference between raw milk and the ultrasonic- and thermosonication- treated samples.

#### Impact of ultrasound on physical properties of milk:

Data in Table (2) show an insignificant differeces (P>0.05) in pH value of control (6.69) to 5 and 15 min (6.70 and 6.70) in ultrasonic- treated (US) and also in thermosonication-treated milks, behaved the same trend as with the acidity.

Regarding the results of the specific gravity of milk sample (Table 2) indicated significant increase ( $P \le 0.05$ ) in control (1.032), compared with the ultrasonic- treated and thermosonication treated samples (1.031, 1.029, 1.031 and 1.027), respectively. Tomislav *et al.* (2012) reported that ultrasonic resulted in no influence on pH value and density.

Table 2. Changes of pH, acidity and specific gravity of milk after the exposure to the ultrasound shock of raw milk (Average±SE of 3 replicates).

Fraction	Control	US		TS		
		5 min	15 min	5 min	15 min	
pH value	6.69±0.19a	6.70±0.13°	6.70±0.04ª	6.65±0.03°	6.58±0.13a	
acidity, %	0.16±0.11a	0.16±0.01a	0.17±0.02a	0.17±0.11	0.18±0.13a	
specific gravity	1.032±0.60°	1.031±0.67	1.029±0.91ab	1.031±0.06	1.027±0.58b	
See legend on Table (1) for details.						

Ultrasonication resulted in release of protein from the micelles to the serum phase (Liu, *et al.*, 2014a). Ultrasonication resulted in release of protein from the micelles to the serum phase. Disruption of casein micelles

was more at higher pH and lower frequency. The effect of ultrasonication treatment (400 w, 24 kHz, 120 Um amplitudes) over heat treatment (63°C for 30 min) on milk, is decreasing pH, increasing in lactic acid content by 0.015% and decreasing density and freezing. All those results came in agreement with Bermúdez-Aguirre and Barbosa-Cánovas (2008).

Due to the effect of thermosoncation (TS) and ultrasonication (US) on the yoghurt starter during fermentation, results in Fig (1) an insignificant decrease (P>0.05) in control compared with US- and TS-treated amples after 30 min period. During fermentation the pH decrease rate on milk samples (US and TS) after exposure to ultrasound shock was slower, compared with the control sample. After 180 min, the pH value in the control, ultrasonication (A, B) and thermosonication (C, D) was 4.90, 5.0, 5.01, 5.21 and 5.20, respectively.

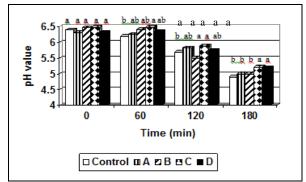


Fig. 1. Changes in pH value during fermentation of samples of milk after the exposure to the ultrasound shock of raw milk (Average±SE of 3 replicates).

Fig. (2) shows that samples were completely different as affected by application of ultrasonic technique. Control sample was of the highest value of CT (62.7 g), while the treated samples with ultrasonic wave were were 36.0, 32.7, 37.7 and 35.5 g, respectively. This might be due to the homogenization effect after the exposure to the ultrasound shock, which agreed with Chandrapala *et al.* (2014) and Nobuyoshi and Etsuzo (2002)

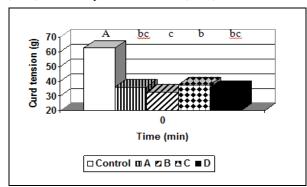


Fig. 2. Changes in curd tension (g) during fermentation of samples of milk after the exposure to the ultrasound shock of raw milk (Average±SE of 3 replicates).

Also, the synersis values (CS) after 30 min of synersis time were 5.16, 7.55, 6.67, 6.58 and 5.13 g/1 g respectively. The CS after 120 min were 8.57, 9.52, 9.37, 9.62 and 8.39 g/15 g respectively (Fig. 3).

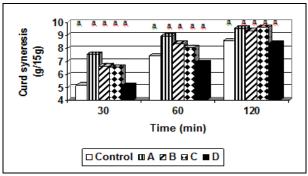


Fig. 3. Changes in curd synersis (CS, g/15 g) during fermentation of samples of milk after the exposure to the ultrasound shock of raw milk (Average±SE of 3 replicates).

Regarding the properties of milk used in cheese making, coagulation time (RCT) characterized with a wide range of variation (Table 3). The highest RCT was of 368.67 sec in control, while the RCT reduced in both of ultrasonication- and thermosonication-treated samples (352.67, 288.05, 315.63 and 338.55 sec, respectively), while treatment of milk with ultrasonication and thermosonication resulted in highest wet curd yield (WCY%) (27.34, 35.0, 31.34, 34.0 and 31.0%, respectively). These results may be due to reduction in the casein micelle size after exposure of milk to ultrasound.

Table 3. Changes in coagulated time (TCR, sec), wet curd yield (WCY), %) and total nitrogen (TN, ng/100 ml) of milk samples after the exposure to the ultrasound shock (Average±SE of 3 replicates).

Fraction	Control	US		TS	
Fraction		5 min	15 min	5 min	15 min
Coagulated time	368.67	352.60	288.05	315.63	338.55
(RCT, sec)	$\pm 4.79^{a}$	$\pm 4.05^{b}$	$\pm 3.46d$	$\pm 3.17^{c}$	$\pm 4.61^{b}$
Wet curd yield	27.34	35.00	31.34	34.00	31.00
(WCY, %)	±0.93°	$\pm 0.28^{a}$	$\pm 0.32^{b}$	$\pm 0.57^{a}$	$\pm 0.28^{b}$
Total nitrogen	0.122	0.122	0.11	0.124	0.120
(TN, ng/100 ml)	$\pm 1.22^{ab}$	±1.22ab	$\pm 1.22^{b}$	$\pm 1.22^{a}$	$\pm 1.22^{ab}$

See legend on Table (1) for details

TN content in the whey changed significant, particularly, in the thermosonication-treated at 5 min (0.124 mg/100 ml). A pronounced increase in this treatment were noticed with the corresponding increase of wet curd yield and decrease in the coagulation, which could be related to losses of casein micelle structure at higher pH aiding Ultrasonication typically induced shear force action in disruption of reassembled casein micelles, which undoubtedly came in harmony with the results revealed with Madadlou *et al.*, 2009, Marchesini *et al.* 2012 and Liu *et al.* 2014b).

From the obtained results, ultrasound could be used in food preservation in combination with other treatments to improve inactivation efficiency. US is applied at low temperature and TS is a combined method of ultrasound and heat. This method showed a great effect on inactivation of microorganisms than heat alone.

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# تأثير الموجات فوق الصوتية على بعض الخصائص الفيزيائية والتكنولوجية للبن الخام منال على نعيم معهد بحوث الانتاج الحيواني

استخدم لبن جاموسي طازج من محطة بحوث الانتاج الحيواني بسخا. وأجريت على العينات سلسلة معاملات بالمعالج المتجانس بالموجات فوق الصوتية وقسمت العينات الى ٤ مجموعات بالاضافة الى الكنترول . المعاملة (أ ، ب) تم تعريضها للموجات فوق الصوتية ٢٠ كيلوهرتز (Ultrasonication) لمدة ٥ و ١٥ دقيقة على حرارة ٢٠ ± ٥ درجة مئوية. والمعاملة (ج ، د) والتي تم معاملاتها بالموجات فوق الصوتية ٢٠ كيلوهرتز على حرارة ٥٠ ± ٥ درجة مئوية (Thermosonication) لمدة ٥ ، ١٥ دقيقة. اظهرت النتائج زيادة في محتوى اللبن الخام من الدهن والبروتين زيادة ضئيلة بالمعاملة (أ ، ب) وكذلك (ج ، د) وايضا كان ملاحظا اختلافات ضئيلة في محتوى اللاكتوز بين اللبن الخام والمعاملات بالموجات الفوق صوتية. وكان هناك اختلافات في قيم الرقم الهيدروجيني والحموضة كما اشارت النتائج ان المعاملة بالموجات الفوق صوتية والمعاملة بالموجات الفوق صوتية تحت حرارة ادت الى انخفاض في قيم الكثافة النوعية العينات ، زادت معدلات طرد الشرش للعينات المعاملة مع انخفاض في قوة الخثرة وذلك للخثرة الحامضية اما الخثرة الانزيمية اظهرت انخفاض في وقت التجين بعد المعاملة بالموجات الفوق صوتية مع زيادة في تصافي الخثرة .