



Effect of Ultrasonication on Microbial, Chemical and Sensory properties of Juices and its Kinetic aspects: A Review

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Abstract : Ultrasound is a novel and innovative technology which is rapidly emerging in food industry. There was an increased consumer demand on new methods of food processing that have a reduced impact on nutritional content and overall food quality. Ultrasonic processing is still infancy and requires a great deal of future research work in order to develop industrial equipment. Ultrasound is found to be an effective method for microbial inactivation and greater efficiency is obtained by combination of ultrasound with heat and pressure. This technique is also used as an analytical technique to provide information about the physiochemical properties of foods. Ultrasound is more beneficial because less processing time, better product quality, less hazards and being eco-friendly. This review summarizes mechanism, application and effects on various parameters of ultrasound in fruit juices.

Key words: Ultrasound, food preservation, microbial inactivation, heat and pressure, food quality.

1. INTRODUCTION

Increasing demands for minimal processed foods which have increased food quality. These demands involve development of innovative technologies with the aim to retain nutritional, non nutritional (bioactive) and sensory characteristics (1, 2). Conventional thermal pasteurization and sterilization techniques inactivate not only microbes and enzymes it also degrade nutritional and organoleptic qualities of foods (3). Non thermal methods such as addition of natural antimicrobial agents in food, high pressure processing, ultrasonication, ozone, pulsed electric field, and ultraviolet is increasingly gaining attention for food processing and preservation. Ultrasonication is one of the emerging techniques which are alternative option to conventional thermal techniques. At mild temperature conditions, ultrasound can pasteurize and preserve foods by inactivating microorganisms and enzymes which can improve food quality in addition to stability and food safety (4). High power ultrasound at lower frequencies (20 to 100 kHz), which is referred to as "power

ultrasound” has the ability to cause cavitations (implosion of gas bubbles) which has uses in food processing to inactivate microorganisms (5). Low power ultrasound at high frequency (1-10MHz) ultrasound used as an analytical technique to obtain information on the physicochemical properties of food such as acidity, firmness, sugar content, ripeness, etc. (6). The low power ultrasound has been used for studying the physiochemical and structural properties of various food products (7).Ultrasound considered to be more advantageous due to its reduced processing time, with lower energy consumption, enhanced quality, reduced chemical and physical hazards, improve the shelf life and being environmental friendly (8). Piyasena *et al.*, 2003 suggested that the future of ultrasonication in the food industry, for purposes of bacterial destruction, is the most promising when ultrasound combined with pressure and heat (3). The ultrasonication is used in food processing like fruit juice processing, milk processing to increase the shelf life. The fruit juices like orange, apple, lettuce, apple cider, tomato are processed by ultrasound. This review will provide a detailed account on the effect of ultrasonication on microbial inactivation, enzyme inactivation, chemical degradation, and sensory properties, including various kinetic models for inactivation studies.

2. MECHANISM AND APPLICATION OF ULTRASONICATION

Ultrasound is defined as sound waves or pressure waves with frequency of 20 KHz to 500 MHz (9).When sound energy passes to the food medium causes cavitation which is the process of generation, growth and collapse of gas bubbles. Due to the violent collapse of bubbles mechanical, thermal, chemical effects are caused. The mechanical effects include collapse pressure, turbulence, and shear stress and chemical effects include free radical production (9, 10).Cavitation bubble collapse due to increase in local pressure(1000 atm)and temperature (5000K) (11). Ultrasound is applied to report constructive effects in food processing such as enhancement of mass transfer, food preservation, support of thermal treatments and food analysis (12). Ultrasound is currently employed in a range of industries, such as surface cleaning, medical scanning ultrasonic therapy, food and beverage technology, nanotechnology, mineral processing, industrial welding, non destructive testing and environmental decontamination applications (13, 14). In food processing ultrasound is widely used for emulsification, extraction, extrusion, defoaming, tenderization of meat, degassing, crystallization, cleaning,leaching, adultration (15). In food preservation ultrasound is used for microbial inactivation, enzyme inactivation, dehydration (1).

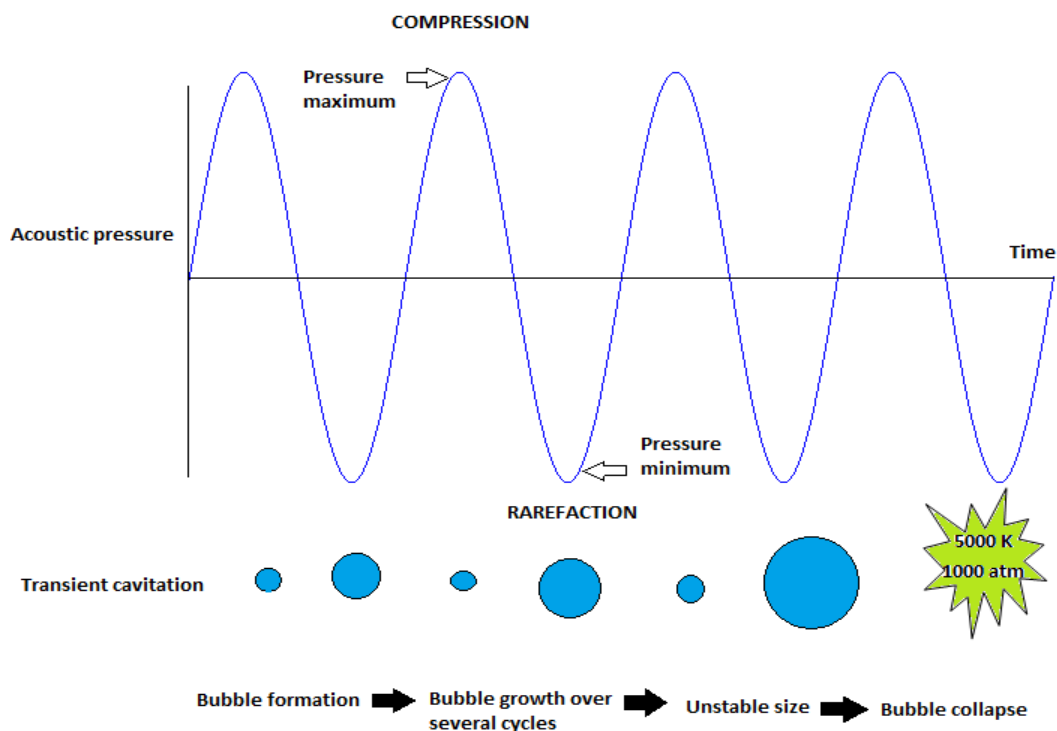


Figure 1 Ultrasonic cavitation

3. EFFECT OF ULTRASONICATION

3.1 MICROBIAL INACTIVATION

Food products are perishable by nature and require protection from spoilage during their Preparation, storage and distribution to give them desired shelf life. The principal reactions that lead to spoilage, are includes physical, chemical, enzymatic and microbiological factors. Microorganisms are the main agents which are responsible for food spoilage and food poisoning and therefore food preservation techniques are targeted towards them (16). Food preservation methods are currently used by the industry on the inhibition of microbial growth or on microbial inactivation. Methods which prevent or slow down microbial growth cannot completely declare food safety (17).

Microbial inactivation by ultrasound produces acoustic cavitation, results in increased permeability of membranes, selectivity loss, cell membrane thinning (18) and DNA damage via free radical production (19). Mechanism of destroying the microbes is due to the shockwaves that create localized mechanical effects with very high temperature and pressure (1). Ultrasonication has achieved the Food and Drug Administration (FDA) requirement of 5-log reduction in microbial survivals (20). Ultrasonication was found to be effective processing for microbial deactivation in *Escherichia coli*, *Listeria monocytogenes* and other spoilage microorganisms (21). Levels of *E. coli* O157:H7 were reduced by 5 log cfu/mL with ultrasound in apple cider and the inactivation of *E. coli* was enhanced using ultrasound at ambient temperatures (22). The study of Abid *et al.*, 2013 on ultrasonication of apple juice did not achieve the complete decontamination and reported that only ultrasonication cannot achieve complete inactivation of microbes (8). The effectiveness of ultrasonication is achieved by combination with heat or pressure or both to get complete sterile product. Sagong *et al.*, 2011 proposed the work on ultrasonication of organic fresh lettuce was reported that the combination of ultrasonication with organic acids (mallic acid, lactic acid, and citric acid) was effective in microbial inactivation (23).

3.2 ENZYME INACTIVATION

Enzymes are responsible for large number of spoilage reactions occurs in food products like enzymatic browning. The enzymes present in foods are needed to inactivate to preserve food and to increase the shelf life (24). The commonly used method for inactivate enzyme is heat treatment but it deteriorate nutrients and causes loss of food quality. So the non thermal technologies are experienced for decrease the enzymatic activities (25). It is well known that ultrasound is in use as a technique for inhibiting the enzyme activity (26). The first enzyme inactivation by ultrasonication is carried out for enzyme pepsin almost 60 years ago and its mechanism of inactivation is defined by cavitation (27). Ultrasound alone is not efficient method due to extent of processing time. Efficient inactivation of enzyme is achieved by combined process like thermosonication or manothermosonication (24). One of the strong heat resistant enzyme in oranges is pectinmethylesterase with help of manothermosonication inactivation of this enzyme is effective (28). Ultrasound produces continuous vibration and collapse of bubbles due to increase of micro temperature and pressure (29). And also by shock waves with strong shear the modification in secondary and tertiary structure of protein is caused due to the breakdown of hydrogen bonds in polypeptide chains, this mechanism because inhibit the activity of enzymes (30). The free radical formation is an important criterion in enzyme inactivation. Peroxidase activity is high in unblanched fruits and vegetables which causes browning and production of off flavors. The activity of peroxidase was minimized by 90% when ultrasonication was applied for 3 hours.

3.3 CHEMICAL PROPERTIES

The chemical properties such as pH, total soluble solids, titratable acidity, ascorbic acid are has effect on ultrasound treatment depending on the process parameters such as amplitude, treatment time, frequency. The work proposed by Tiwari *et al.*, (2009), has no significant effects are observed on both titratable acidity and total soluble solids (° Brix values) (31). The pH has a small considerable increase during storage. Ascorbic acid was decreased during the storage, higher ascorbic acid retention was observed in ultrasound treated sample on treatment time for 10 mins with amplitude level of 100%. Cortes *et al.*, (2008) reported that pH was increased during refrigerated storage of orange juice treated with high intensity pulsed electric field (32). Ascorbic acid degradation was less for sonicated juices compared to thermally processed juices (33). In ultrasonic process samples the ascorbic acid degradation is related with oxidation reaction, promoted by free radicals generated by cavitation (34).

Cruz-Cansino *et al.*, (2016) reported from their work that no detectable levels of *E. coli* in cactus pear juice for 2 days with no effect on pH, titratable and total soluble solids on ultrasound treatment at 90% amplitude for 5 min and after 5 days significant changes were observed on the above noted parameters (35). The compounds like sugars, phenolic compounds and organic acids are released during the ultrasound treatment. For example the increase in titratable acidity after treatment and storage is due to the release of citric acid during ultrasound treatment. There was no significant difference in pH, soluble solids, acidity in sonicated apple juice (8). The same results are observed by Bhat *et al.*, (2011) on kasturi lime juice except °brix value was decreased after the sonication treatment (36).

3.4 SENSORY EVALUATION

The significant decrease on sensory parameters (color, odour, taste, and aroma) was observed on ultrasonically treated apple juice and nectar compared with untreated samples, but there was no rejection of the product was found on the work done by (37). The changes in color and taste of fruit juices are due to cavitation produced during the ultrasound treatment. Vercet *et al.*, (1998) suggest that metallic taste could appear due to changes caused by free radicals produced during manothermosonication of apple juice and cranberry (38). Ultrasound of fruit juices show minimal effect on color change compared to conventional heat treatment. The positive effect of ultrasound is efficient removal of dissolved oxygen from the raw juice compared with conventional process (39). The changes in odour and taste of sonicated juice are due to the formation of various compounds and oxidation which interacts with free radicals formed.

3.5 NUTRITIONAL PROPERTIES

Ascorbic acid is considered as an indicator of the nutritional quality of juices (40). It is an important nutrient that shows antioxidant ability and provides the protection against free radicals (41). Fruit juices are generally rich in simple carbohydrates and sugars are the main components in fruit juices. Around 80% of the total soluble solids are composed of sugars which are accountable for the sweetness of the juice (42). The sugar profile and ratios of specific sugars are considered as an indicator for determining the effect sterilization techniques on fruit juices. Rocha *et al.*, 2003 observed that with an increase in the processing and storage time of juice, a significant decrease in the glucose content was observed, which can be attributed to the use of sugars as energy sources and substrates in metabolic processes of microbes (43). Khandpur and Parag, (2015) had a work on ultrasound treatment of various juices shown a result that application of ultrasound treatment had preserved the total phenolic and antioxidant levels of fruit juices and maintain the carbohydrate profile in better manner during the storage as compared to the thermally processed juices (44).

4. ULTRASONICATION EQUIPEMENT OPERATION

The three crucial parts of ultrasound equipment are generator, transducer, and coupler (45) Ultrasonic generators transform electrical energy into ultrasound energy (mechanical energy) passing through a transducer. The frequency of ultrasonicator ranges from 20 KHz to 40 KHz. By means of piezoelectric effect ultrasonic waves are generally produced by ceramic crystal enclosed within a transducer that is energized by electrical energy. Povey and Mason, (1998) classified different types of transducer such as magnetostrictive, liquid-driven, and piezoelectric transducers (46). In which piezoelectric transducers are 95% energy efficient and commonly found in probe and bath type sonicators. The probe or horn type and bath type sonicators operated in batch process. The frequency and power ranges were dissimilar for both ultrasonicators.

Ultrasonic cleaning or bath system consists of stainless steel jacketed vessel, sonication bath, time, and temperature control system. An ultrasound generating transducer built inside the chamber, or lowered into the fluid, produces ultrasonic waves in the fluid by changing size in concert with an electrical signal oscillating at ultrasonic frequency. An aqueous (water) or organic solvent, depending on the application was used as an ultrasonic waves transfer medium. In probe type ultrasonicator the probe transfers ultrasonic vibrating energy in to the liquid foods. During operation, the probes tip longitudinally expands and contracts this produces cavitation.



Figure 2 Probe type ultrasonicator

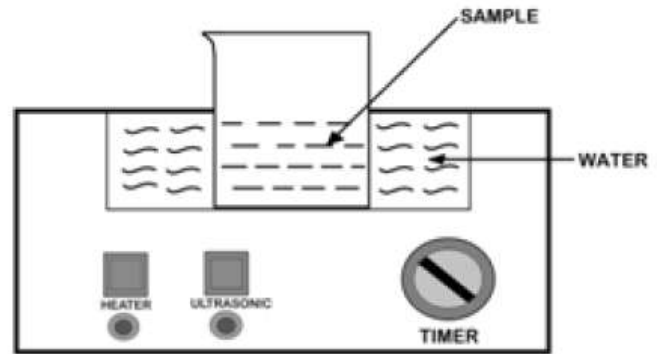


Figure 3 Ultrasonic bath

5. INACTIVATION KINETICS

To ensure the food safety by food preservation process kinetic parameters and models are used. These parameters used to analyze and report the reduction of a microbial population, process parameters include coefficients which are experimentally determined from microbial reduction kinetics and from expressions based on chemical reaction kinetics. These parameters and models used to compare the processing of different technologies on microbial population reduction. The purpose of this section is to review various models for microbial inactivation.

Table 1 Various primary models taken from (16).

Models	Author Name	Equation
Gompertz model	(47)	$\log N_t = A + C \times \exp [-\exp [-B (t-M)]]$
Modified Gompertz	(48)	$y = a. \exp[-\exp (b-ct)]$
Logistic model	(49)	$y = a [1 + \exp(b- cx)]$
Baranyi model	(50)	$y(t) = y_0 + \mu_{\max} F(t) - \left[\ln \left(1 + \frac{e^{\mu_{\max} F(t)} - 1}{e^{y_{\max} - y_0}} \right) \right]$ <p>Where,</p> $F(t) = t + \frac{1}{v} \ln (e^{-vt} + e^{-h_0} + e^{-vt-h_0})$
First-order monod model	(51)	$\mu = \frac{\mu_m S}{K_s + S}$
Modified monod model	(52)	$\mu = \mu_m \times \frac{\alpha(P' - P)}{P'(\alpha - P)}$

PARAMETERS EXPLAINATION

- N_t - the cell density at time t , A - the lower asymptotic line of the growth curve as t , C - the difference between the upper asymptotic line of the growth curve (maximum population level), B the relative maximum growth rate (h) at time M , M - the time at which the growth rate is maximum.
- $y(t)$ - the \log_e (CFU ml^{-1}) of cell concentration at time t , y_0 - the initial cell concentration in \log_e (CFU ml^{-1}) units, y_{max} - the maximum cell concentration in \log_e (CFU ml^{-1}) units, μ_{max} - the maximum specific growth rate in terms of \log_e (CFU ml^{-1}), v - the rate of increase of the limiting substrate, assumed to be equal to μ_{max} , h_0 - equal to $\mu_{max} \times \lambda$ and λ is the lag-phase duration in h .
- μ and μ_m are the actual and the maximum specific growth rates (h^{-1}), S is the rate limiting substrate (nutrient) concentration ($gS\ l^{-1}$), K_s is the saturation or half-rate constant ($g\ l^{-1}$) and X is the biomass concentration ($gX\ l^{-1}$).
- P is the salt concentration (millimolar), μ_m is the specific growth rate at $P=0$, P' is the MIC, and α is a shape parameter.

6. CONCLUSION

Ultrasound finds a diverse application in science and food technology in the future of food industry. The use of ultrasound on its own in the food industry for microbial and enzyme inactivation is currently unfeasible, however the experiments in which ultrasound has been coupled with heat and pressure have been very successful. Conventional thermal processing results in destruction of nutrients and flavors of the product and it have low energy efficiency. To overcome these disadvantages ultrasonication was used as a novel technology. It has advantages of minimizing flavor loss, increasing homogeneity, saving energy, high productivity, enhanced quality, reduced chemical and physical hazards and is environmentally friendly. Several research works had reviewed on ultrasonication of juices in this work.

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