## ULTRASOUND ASSISTED THERMAL PROCESSING FOR ENERGY-SAVING AND MILD PRESERVATION OF LIQUID FOOD

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## ABSTRACT

In the present study a continuously working pilot plant scale prototype has been used to evaluate the effects of continuous flow ultrasound-temperature treatment for bacterial decontamination of model suspensions and various liquid food systems such as for milk, fruit and vegetable juices. The inactivation of *Escherichia coli K12 DH 5 a* and *Lactobacillus acidophilus* has been examined. Moreover treated juices have also been investigated for damage caused by heat or ultrasound induced degradation of organoleptic and nutritional properties after treatment and storage. In particular changes in colour and destruction of heat labile and slightly oxidizable Lascorbic acid has received special attention. The achieved results were assessed with respect to the energy requirement and compared with those obtained using a conventional heating method having similar processing conditions.

#### INTRODUCTION

The concept of ultrasound assisted thermal processing, which makes use of the synergy between ultrasound and heat for bacterial inactivation, has proven to be of potential interest in food preservation. First introduced by Ordonez (1987), it's applicability was predicted for support of traditional methods using elevated temperature. Suited to pumpable liquids which could be processed in a continuous flow arrangement with clean or aseptic filling (Earnshaw, 1998) it was proposed for the reduction of process temperatures and/or process times of pasteurisation or sterilisation processes to achieve the same lethality values (Mason, 1996). The reduction of the temperature applied and/or of the time of treatment should result in a lower energy requirement and, above all, in a reduced detrimental effect on food.

The majority of studies about the bactericidal efficacy of ultrasound and ultrasound assisted thermal processing were investigated in laboratory scale batch-type ultrasonic reactors. Detailed experience in continuous flow in particular with reference to energy and food quality assessment are very scarce. During the last few years some studies appeared where single or combined effects of ultrasound were investigated in continuous flow (Villamiel et al. 1999, 2000 and Williams et al. 1999). The experimental results achieved in these studies lead to further investigations of a combination preservation process with regard to its total energy consumption and to its impact on product quality.

In order to compare the ultrasound assisted treatment with conventional heating for bacterial inactivation various non-linear effects associated with intense sound fields had to be considered. The ultrasound generated acoustic streaming taking into account the viscosity of the

treatment medium is required for that in addition to the temperature effect as a result of the specific absorption of acoustic energy.

For considering the temperature effect the temperature dependent process lethality was calculated from each treatment. The theory includes the integration of the mean measured timetemperature profiles of the investigated processes by using the z factor of the test organisms after expression of the decimal reduction time as an explicit function of temperature. The utilization of the basic principles of thermal process calculations gives the permission to consider the temperature effect first in it's strongest simplified form. Precisely taken the approach/evaluation has proceed on the assumption of a radial uniform temperature and velocity distribution resulting from a radial mixed product flow (perfectly mixed flow) and the expectation that the internal heat generation during sonication was homogeneous over the radius due to a volumetric energy dissipation.

#### MATERIAL AND METHODS

#### Laboratory scale batch-type treatments

Experiments were performed using *Escherichia coli* K12 DH 5  $\alpha$  (Hygiene Institut Hamburg, Germany) and *Lactobacillus acidophilus* ATCC 4356 (DSM, Braunschweig, Germany), which were chosen with regard to gram behaviour, sonication cell sensitivity and importance as indicator organisms for an undesirable contamination of the investigated food products.

To control the actual temperature during conventional and ultrasound assisted heating 9 ml of sample medium were placed in a thin glass vessel and heated by a thermostatically controlled bath (Haake, Model DC5, Karlsruhe, Germany) until steady state conditions. Inoculation of 1 ml cell suspension was done when the medium (9 ml treated volume) had reached the temperature selected for the treatment. In the course of the ultrasound assisted treatment the final temperature were adjusted during sonication. Ultrasound at 20 kHz and 50, 110 or 160  $\mu$ m of wave amplitude was applied to the medium using a Sonopuls HD 2070 homogenizer (BANDELIN electronic & Co. KG, Berlin, Germany) equipped with a SH 70 G horn and a KE 76 tip.

Pilot-scale treatment in continuous-flow

About 60 I freshly inoculated sample medium were treated after 1 h adaptation time at 20°C for the test organisms. The experimental setup employed was the same as that used in previous studies and was already described in detail (Zenker et al., 1999; Zenker et al., 2000). The sample medium was indirect heated by a plate heat exchanger and than conveyed through the holding section (d = 20 mm, l = 1500 mm) into the cylindrical sonication cell (inside: d = 19.8 mm, h = 380 mm). It was than chilled by counter-current cooling after passing a pressure control valve which allowed the regulation of the flow rate and the operation in the range of 3.5 - 4.5 bar systempressure (absolut).

During ultrasound assisted experiments irradiation was carried out using an industrial ultrasound processor (Model UIP 1000, Dr. Hielscher GmbH & CO KG, Teltow, Germany) attached at the head of the vertical installed sonication cell. The acoustic source creates a high frequency oscillation of 19.3 kHz at amplitudes between 45-55  $\mu$ m within the medium streamed from below. It was equipped with a specially created cone tip with 8.32 cm<sup>2</sup> front area and calibrated for frequency resonance in the mentioned pressure range to maintain a maximal power input of approximately 700-800 W.

The pilot plant unit was purposely provided with a equipment suited for a permanent monitoring and data acquisition of mass-flow, processing pressure and -temperature. Among others it allowed the control and the determination of mean time-temperature-profiles at any operation conditions. To achieve different process intensities the conventional and the ultrasound assisted heat treatment were performed with gradual increase of the heat flow rate. Other critical process parameters such as pressure, mass-flow and effective ultrasound output were kept constant.

#### Determination of L+ ascorbic acid

Ascorbic acid levels were determined by using the colourimetric assay from Boehringer (No. 409 677; Manheim, Germany) and carried out by adding 0.1 ml of the filtered sample () into cuvettes at room temperature, adding 1 ml of 0.75 mM 3-(4.5-dimethylthiaolyl-2)-2.5-diphenyltetrazolium bromide solution (MTT) and 1.5 ml destilled water. In the presence of 0.1 ml

15 mM 5-methylphenazine methosulfate at pH 3.5, the MTT was reduced by the ascorbic acid. The resulting formazan was photometric detected by absorbance at 578 nm after 15 min incubation at 37°C.

#### Colour measurement

Colour after treatment or during storage period was instrumental determined using the CR 200 model Minolta Chromameter (Minolta Camera Co., Osaka, Japan) with triplicate measurements for each determination. The CIE L\*a\*b\* colour notation system was applied to measure the parameters L\*, a\*, b\* where L\* indicates the lightness, a\* means the colour axis from red to green and b\* the yellow-blue axis.

### **RESULTS AND DISCUSSION**

#### Laboratory scale batch-type experiments

The destruction kinetics of the test organisms for both conventional and ultrasound assisted heating result in a first order reaction, which enabled the determination of D and z by linear regression as well as the evaluation of the inactivation results by using a simple time law. In addition the kinetics for sonication at ambient temperature level showed the same characteristics.

Based on the batch mode kinetics data (Table 1) a significant enhanced effect on *E. coli and L. acidophilus* destruction as result of the ultrasound assisted thermal processing has been detected. In buffer media at a temperature of 60°C the combined process reduced the D value of *E. coli* by 72.7% and *L. acidophilus* by 38.6% as compared to the D value of the heat treatment alone and by 71.6% or 70.1% (*E. coli*) as well as by 34.2% (*L. acidophilus*) when tested in carrot juice, milk or orange juice. These percentages represent for *E. coli* at least a 3.3 fold and for *L. acidophilus* a 1.5 fold reduction in time or at the same Dvalue a 1.18 fold (*E. coli*) or a 1.02 fold (*L. acidophilus*) reduction in temperature to degrade the cell count about one log cycle.

Table 1. Kinetic factors D and z for *E. coli DH* 5 a and *L. acidophilus* inactivation by thermal and ultrasound assisted thermal treatment (sound wave amplitude 110  $\mu$ m) in the batch-type reactor.

organism	E. coli K 12 DH 5 a						L. acidophilus			
medium	phosphate buffer (ph 7.0)		carrot juice (pH 5.9)		UHT-milk (pH 6.7)		phosphate buffer (pH 7.0)		orange juice (pH 3.7)	
treatment	Т	TS	Т	TS	Т	TS	т	TS	Т	т
D <sub>60</sub> -value [s]	84.6	23.1	84.3	23.9	77.0	23.0	70.5	43.3	47.3	31
z₄8-68 -value [°C]	6.9	16.3	8.7	15.3	7.1	13.6	6.1	7.5	5.5	7.

T: conventional heating, TS: ultrasound assisted treatment

#### Pilot-scale experiments in continuous-flow

#### Microbial inactivation study

During ultrasound assisted heating performed in continuous-flow the bacterial inactivation was consistently higher than in the conventional heating process in the same way as in the laboratory scale batch-type experiments. This fact was recognized, as for the secure proof the results of microbial destruction have been compared with the integrated lethal value of heat (F-value) calculated for each treatment (Fig. 1).

The impact of the combination treatment on the killing rate is evident when a reference process at a constant temperature level is used. With *Escherichia coli K 12 DH 5 a* and *Lactobacillus acidophilus* in phosphate buffer a 3 fold and 1.5 fold higher inactivation was found.

However the effect was detectable up to a mass-flow of 26 l/h and reproducible within the bounds of the carried out evaluation. Besides the determination of z, which has be done in the previous batch-type experiments, it included the measurement of the mean time-temperature

profile of the pilot plant unit. With it the computation of F-value based on the ideal assumption of a uniform radial temperature and velocity distribution in other words of plug flow or perfectly mixed flow conditions.



Fig. 1. Inactivation of *E. coli DH* 5 a(A) and *L. acidophilus* (**B**) after conventional (o) and ultrasound assisted beating (b) within the pilot plant unit. Mean residence time

and ultrasound assisted heating (•) within the pilot plant unit. Mean residence time within the ultrasound reaction chamber [s]: ca. 60; specific sound energy input [kJ/kg]: 96, mass flow [l/h]: 26, treatment medium: phosphate buffer (pH 7), standard deviation (single random sample, six repetition measurements) [log N/No]: 0.59 (*E.coli*) and 0.55 (*L. acidophilus*).

# Energy balancing

In order to quantify the combined ultrasound and heat treatment with regard to its potential in reducing the total process costs, balancing of the total energy input has been applied to both processes. The total energy consumption required for heating and for the acoustic irradiation was compared (Fig. 2).



Fig. 2. Comparison of energy consumption for *E. coli DH 5 a* (A) and *L. acidophilus* (B) inactivation by thermal and ultrasound assisted thermal treatment.

For the derivation of the enthalpy balances thermal heat recovery rates of 94% (indirect heating) has been considered. Although microbial destruction occurred on a significant lower temperature level in the case of the combined process a reduced net energy input could not verified. This has to be attributed to the substantial electric energy loss of ultrasound generation. As presented in Figure 2 the electric consumption significantly exceeded the level of heat energy reduction. Improved results were obtained by applying the same energy balancing procedure to the sterilisation process.

#### Food quality assessment

The investigation of the impact of the storage conditions on the colour of orange juice indicate no detrimental effect in the case of the combined ultrasound application (Fig. 3). Regarding the brightness (L\*-value) a slight improvement was observed which may be due to the lower thermal processing intensity of the ultrasound combined treatment. Moreover, the slower decrease of L\* produced a significant difference of 1.5 after 35 days. The other characteristic parameters of the colour measurement (a\*-, and b\*-value) indicated no difference between the combined process and conventional heating.

To check the influence of the processes on the stability of nutritional valuable compounds Lascorbic acid has been chosen because of its sensitivity regarding oxidative decomposition. Figure 4 show the loss of ascorbic acid occurs during storage of non-, combined- and heat processed orange juice. At an ambient temperature of approx. 20°C and storage in glass bottles a relatively high monthly loss of 3.8-12.3 % was found. The quantity varies, however, due to the applied treatment. In conventionally heated juice a higher decay in significant quantity of 4.7% was detected.



Fig. 3. Results of colour measurement during storage of combined ultrasonic and conventional heated orange juice. (L\*-lightness, a\*-red/green, b\*-yellow/blue).

## CONCLUSION

Preservation of model suspension and fruit- or vegetable juices yielded that a combined processing of heat and ultrasound can be used to reduce the required heat intensity compared to the conventional thermal process. In continuous flow with a rate of 26 l/h a slight retention of the colour was observed in either apple and orange juice. Additionally a significantly lower reduction of native L(+)-Ascorbic acid in orange juice was detected.

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Fig. 4. Degradation of native ascorbic acid in orange juice stored in bottles at 20°C in darkness. In the study the rehydrated juice (11.2°Bx, pH 3.7) was pasteurized at  $P_{60} = 240$  seconds (thermal) and  $P_{60} =$ 150 seconds (ultrasound assisted thermal treatment) with regard to the *L. acidophilus* inactivation.

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