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Ultrasonic characterization of yogurt fermentation process

D. Izbaim[a], B. Faiz[a], A. Moudden[b], M. Malainine[a] and I. Aboudaoud[a]

[a]Faculté des sciences d'Agadir - Université Ibn Zohr, Département de physique,Faculté des sciences d'Agadir, B.P 8106 Agadir, Maroc, 80000 Agadir, Morocco
[b]Ecole Supérieure de Technologie d'Agadir, B.P 33/S Agadir, Maroc, 80000 Agadir, Morocco
dizbaim@gmail.com

The objective of this work is to characterize the fermentation of yogurt based on an ultrasonic technique. Conventionally, the acidity of the yogurt is measured by a pH meter to determine the progress of fermentation. However, the pH meter should be cleaned and calibrated for each measurement and, therefore, this method is not practical. In this regard, ultrasonic techniques are fast, non-invasive and inexpensive. The measurement of ultrasonic parameters such as amplitude and time-of-flight of the echoes, backscattered by samples of yogurt, were able to provide information on the phases of the fermentation and on the state of yogurt over time. The major contribution of this work is the detection with high precision the moment when the fermentation process should end to stop the growth of bacteria.

1 Introduction

Yoghurt is produced through lactic acid fermentation by adding lactic bacteria to milk. This involves the production of lactic acid by the decomposition of glucose during fermentation [1]. Milk coagulation is induced by the solidification of the protein in the milk by this acid.

Lactic acid bacteria are widely used in the food industry to manufacture several fermented foods such as dairy, meat or diverse vegetable products. Lactic acid is used in chemical, cosmetic, textile and pharmaceutical industries as an acidifier, preservative, green solvent, for the production of emulsifying agents and, recently, for the synthesis of biodegradable polymers for medical applications [2].

The quality of the yoghurt can be examined from different aspects by using different methods, reading its chemical, physical, microbiological and nutritive characteristics [3]. Conventionally, the yoghurt acidity is measured by a pH meter to determine the progress of fermentation. Nevertheless, a pH meter requires calibration, and therefore, this method is inconvenient and time-consuming. In this respect, ultrasonic techniques are fast, non-invasive and inexpensive.

Measurement techniques based on the ultrasonic wave propagation analysis have definitively contributed to the recent development of biology and chemistry. Ultrasonic wave propagation characteristics depend on the medium properties, and therefore, ultrasonic techniques are often used for material characterization. Many studies have been conducted to assess the composition and structure of different types of food products using ultrasound [4]. In the dairy research, milk coagulation time using a pulse–echo immersion technique was measured [5]. Recently, ultrasonics have been used for monitoring yoghurt fermentation process [6], and for the detection of microbial contamination in closed UHT milk packs [7], and also to assess the texture of cheese [8].

Though the application of ultrasonic techniques for the analysis and characterization of different materials and processes is well stated, analytic instruments for biochemistry and microbiology based on ultrasound or other kinds of mechanical waves are not usual. However, these techniques may give a new aspect to biochemical processes despite the special features required. Measurement of ultrasonic parameters such as amplitude and velocity through a given medium can provide information about the changes and variations taking place as a function of time.

The objective of this work is to characterize yoghurt fermentation basing on the detection of amplitude and time-of-flight of ultrasonic waves in the sample under study. The main features of this ultrasonic device are analysed in this work, and some measurements and preliminary results are shown.

2 Materials and methods

The experimental setup (figure 1) used for the experiments consisted of ultrasonic transducer (5 Mhz, 0.5 in. crystal diameter, A309S-SU Model, Panametrics, Olympus), attached to a cubic container (50 x 50 x 50 mm) where the milk sample was placed. The container was introduced into a temperature-controlled bath to maintain the sample temperature and the oil was moderately stirred to avoid formation of bubbles. The transducer was linked to a pulser–receiver (Sofranel Model 5073PR, Sofranel Instruments) which sent the electrical signal to a digital storage oscilloscope (LeCroy 9310M, LeCroy Cor).

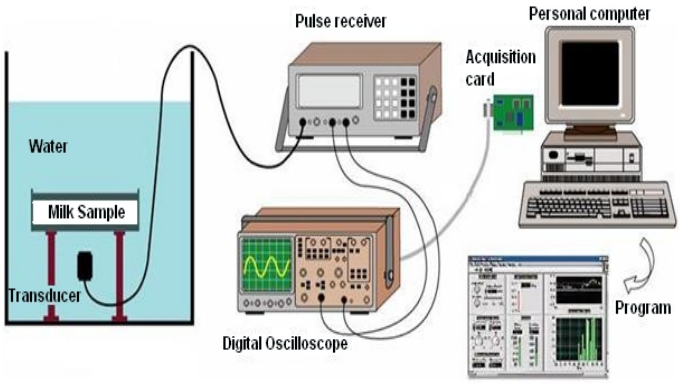


Figure 1: Experimental set-up

The ultrasonic impulse is propagated in water and crosses the sample contained in the vessel before reflecting on a Plexiglas surface of plate (figure 2). Figure 3 shows the experimental signal composed of echoes A1 to A4.

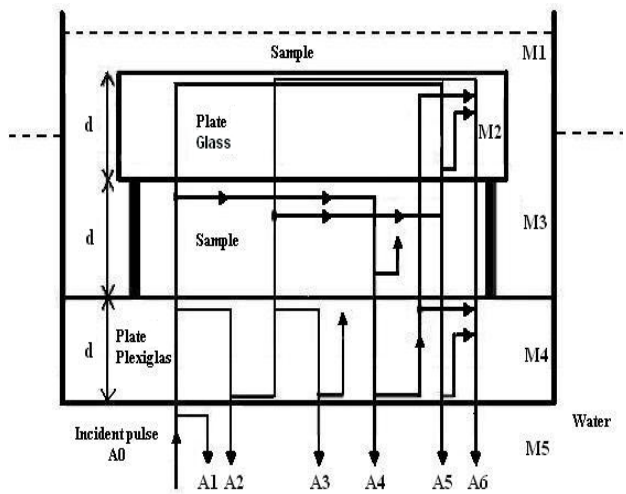
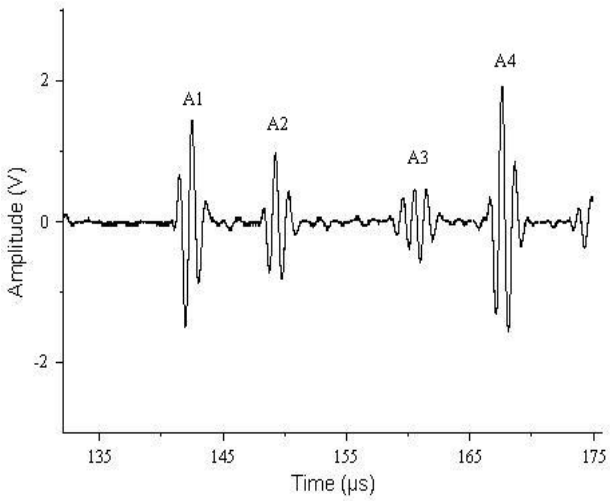


Figure 2: Schematic of echoes reflected by the sample (A1 to A6 are observed echoes reflected from the interfaces between media Mi).



Figurre 3: Typical waveform of the reflected ultrasonic signal with the 5 MHz transducer.

For ultrasonic velocity measurement, eight signal acquisitions were taken and averaged. It can be written as

$$C_{sample} = 2\omega L / \varphi_{A4} - \varphi_{A2} \quad \text{with} \quad \omega = 2\pi\nu \qquad (1)$$

To obtain the phase experimentally, the FFT of signals A2 and A4 are calculated (Bakkali and al, 2001).

The attenuation coefficient α was computed by fitting the experimental data to Eq.

$$\ln A = \ln A_0 - \alpha d \qquad (2)$$

Where d is the distance travelled by the wave, A_0 is the initial amplitude of the signal measured as the peak-to-peak voltage and A is the amplitude of the signal at distance d. Eight ultrasonic echoes were considered to compute attenuation.

3 Results and discussion

We begin the control with the acquisition of the signals represented on the screen of the oscilloscope by the mean of an application under the programming language LabVIEW. The realized program allows to specify the number of acquisitions wanted during the experience and the time separating each one from the other. This program gets each time 50 signals and produces an averaging to neutralize the noise signals. The resulting signal represents for the user a single acquisition (Figure 4).

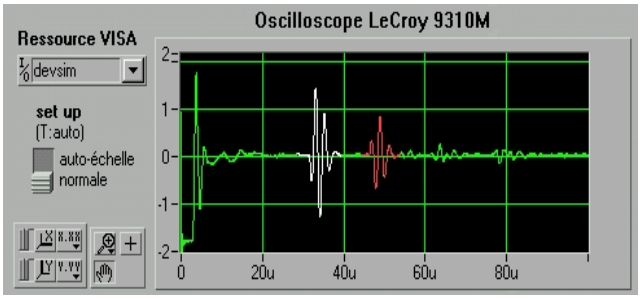


Figure 4: Time domain signal captured from the oscilloscope

Then, the user has the possibility of choosing the first time the echo manually which will take a different color afterward. The first echo (in White) represents the energy returned by the interface Plexiglas / milk and the second (in Red) represents that of the interface Milk / Glass. The program separates then the echo A4 through a temporal window and nullify all that is outside of the window (Figure 5).

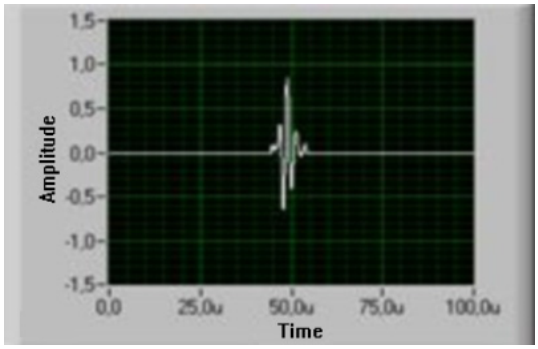


Figure 5: Separation of the echo A4

During the fermentation, the locations of the echos with regard to time (X axis) change and it is due to the change in the crossed middle nature. We calculated the amplitude and the time-of-flight of the echo A4 during the fermentation which lasts 4 hours (Figures 6 and 7). The figure 8 illustrates the behavior of the fermentation in degree of pH.

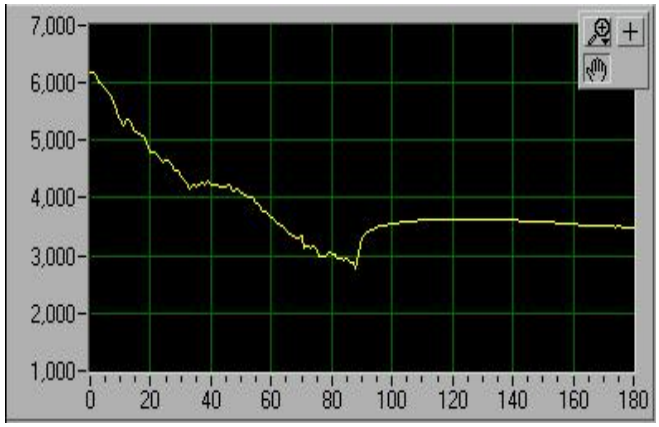


Figure 6: The amplitude of the echo A4 during fermentation

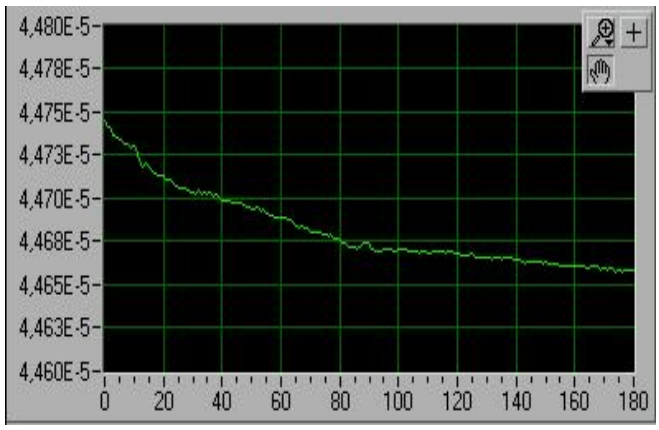


Figure 7: The time of flight of the echo A4 during fermentation

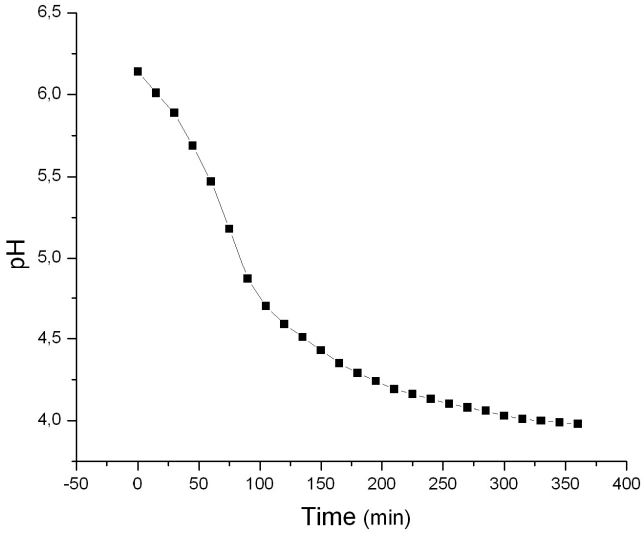


Figure 8: The evolution of the pH degree during the fermentation

By comparing figures 6 and 8, we observe that when the pH is equal to 4.6 value, the amplitude of A4 stabilizes and reaches a maximum after 125 minutes of the beginning of fermentation. However the time-of-flight shows only a small change in its evolution which corresponds to a trough in the curve of the amplitude at 90 minutes.

The review of Figure 7 shows that the evolution of time-of-flight decreases in several steps during the fermentation of milk . This decrease, which means an increase in the ultrasonic velocity, may be interpreted as a change in the mechanical strength of the medium, because the ultrasonic velocity is directly related to the elasticity through the following relationship:

$$V\ (m/s) = (E/\rho)^{1/2} \cdot f(\sigma) \qquad (3)$$

E is the elastic modulus, ρ is the density and σ the Poisson's ration of the medium.

Although its development is not as eloquent, the time of flight indicates the four phases in the evolution of the amplitude. By comparing the curves of the amplitude and time of flight (Figure 9), we find that in the first and second phase, the time of flight regularly follows the amplitude. In the third phase, the flight time reached a plateau, so that later her moderately declining trend in the fourth phase showing increasing strength of the medium.

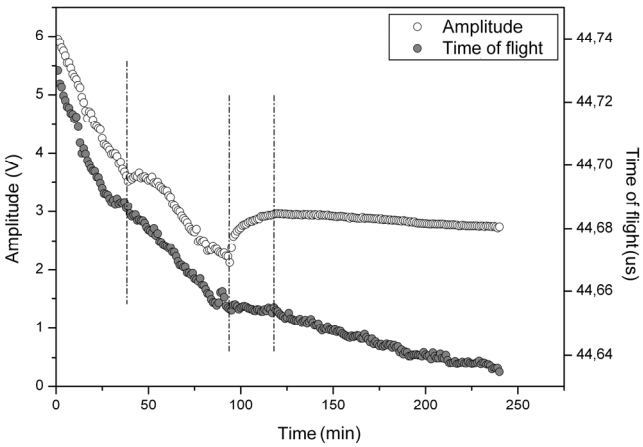


Figure 9. Comparison of evolutions of time of flight and amplitude

Since time of flight marks the four phases found in the amplitude, we thencombined their curves in order to have an evolution curve that represents them andthat includes their information. We proceeded as follows:

$$f(t) = \text{Amplitude} + (100.dT) \qquad (4)$$

dT is the variation with time of flight.

Multiplication by 100 is made for the variation of time-of-flight, which is of the order of 10^{-2} μ s, that affects the amplitude by their addition.

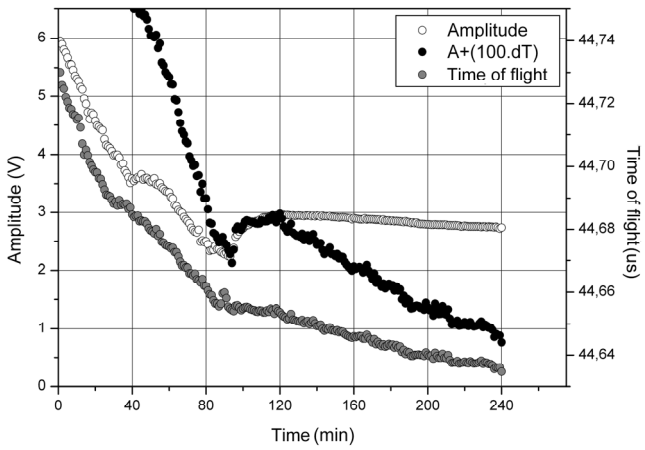


Figure 10. Comparison of evolutions of time of flight and amplitude with the evolution curve resulting from their combination.

Figure 10 compares the resulting curve with those of the amplitude and time-of-flight. The new curve shows the evolution of the viscoelastic parameters of yogurt. We note that this evolution shows more clearly the points pH = 4.9 / 94min and pH= 4.6 / 120min, which correspond to the beginning of the formation of three-dimensional network of yogurt and its stabilization. Point pH = 4.6 is easy to detectin this development than in the amplitude. Using this evolution in the automatic real-time, we will have a maximum delay of 3 minutes instead of 5 minutes when usingthe evolution of the amplitude.

To detect automatically and in real time the point pH=4.6, we treated the amplitude curve mathematically by applying the function PolyFit to eliminate the small fluctuations (Figure 11), then we apply the Derived function to the resultant curve. When the amplitude reaches a maximum (125 minutes), the derivative nullifies (Figure 12) and the program shows a message which informs the user that the fermentation should stop.

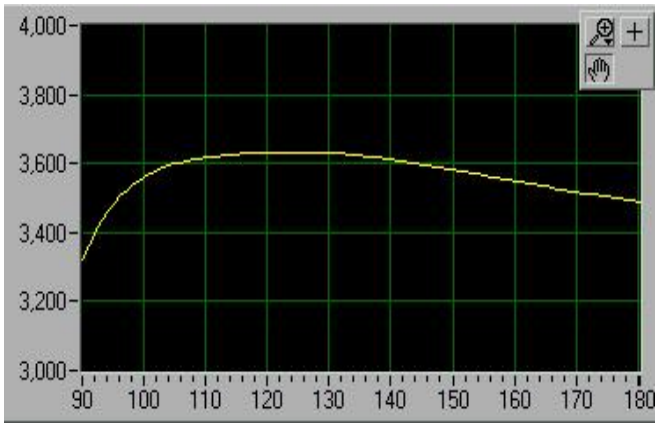


Figure 11: The Polyfit function of the echo A4 amplitude

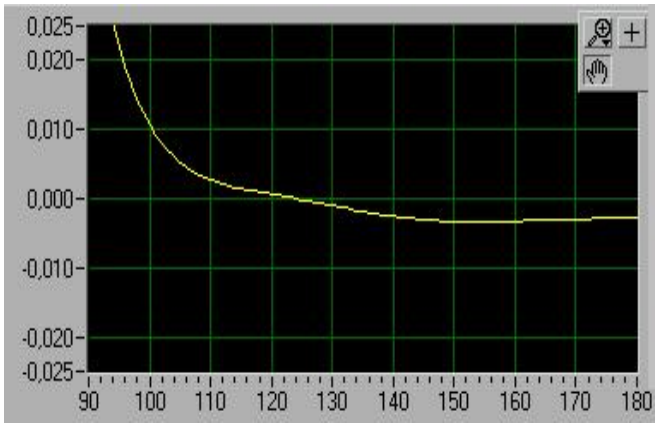


Figure 12: The derivative function of the Polyfit curve

4 Conclusion

The found results show that the ultrasound technique can be an alternative to using pH measure in the dairy industry. This technique is less expensive and requires no expertise, especially as she realizes a real time control by means of a program.

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