Use of a Model Solution for the Evaluation of Heat Damage in Milk Treated in an Ultrahigh-Temperature Heat Exchanger†

Carlo Pompei* and Margherita Rossi

Dipartimento di Scienze e Tecnologie Alimentari e Microbiologiche, University of Milan, via Celoria 2, 20133 Milano, Italy

Heat damage in milk can be determined by measuring either reaction products, whose formation is time–temperature dependent, or the loss of thermolabile constituents contained in raw milk. The purpose of this study was to find a relationship between heat damage in milk and the performances of a 10% sucrose solution at pH 3 used as a model solution. Heat treatments were performed on a laboratory indirect heat exchanger especially assembled. The investigation was based on the experimental determination of the time–temperature profiles in the plant, the kinetics of sucrose inversion in the model solution, and the reaction kinetics of some indices of heat damage in milk, such as the formation of lactulose and the loss of α-lactalbumin. On the basis of the percentage of noninverted sucrose remaining in the model solution, a calculation procedure is proposed for checking the performance of an ultrahigh-temperature plant with reference to heat damage and the bacteriological reduction.

INTRODUCTION

The heat treatments that milk undergoes during sterilization cause chemical changes which can affect its nutritional and sensorial quality. The extent of such heat damage reactions has been widely investigated, and the kinetics of a number of them are already known (Andrews, 1985; Fink and Kessler, 1986; Kessler and Fink, 1986; Dannenberg and Kessler, 1988; Mauri et al., 1989). Knowledge of these kinetics enables maximization of the sterilizing effect and minimization of the heat damage to optimize the milk sterilization processes. Mild technologies, such as HTST and UHT treatments of milk, are based on the above principle. For continuous processes, sterilization heat treatments are customarily based on time and temperature in the holding section. The role of the heating and cooling phases is usually neglected, although these can considerably increase the heat damage to the product.

Frequently, even manufacturers know only roughly the time–temperature profile of the sterilization plant they produce. Actually, the experimental measure of the time–temperature profile in plants for continuous-flow processes is objectively troublesome. This is partly due to the difficulty of knowing the velocity distribution of the fluid particles in each section of the plant. Moreover, users often adjust to their needs the standard operative conditions of the plant without evaluating the effect on the quality factors of the product.

Heat damage in milk can be determined by measuring either reaction products, whose formation is time-temperature dependent, such as lactulose, (hydroxymethyl)-furfural, and furosine (Geier and Klostermeyer, 1983; Keeney and Bassette, 1959; Erbersdobler et al., 1987), or the loss of thermolabile constituents, contained in raw milk, such as thiamin or whey proteins (Burton, 1984). Among the latter, α-lactalbumin (α-LA) is the most heat resistant and could be used to determine the heat damage in sterilized milk (Resmini, 1989).

The literature on some of these reactions reports the value of activation energy (E_a) and, related to this, the value of z (°C). The latter represents the increase in temperature that causes a 10-fold increase in the reaction rate. If both the value of z and the exact time–temperature profile of the fluid in the plant are known, the severity of heat damage, in relation to a well-defined chemical or physical change occurring in milk, can be expressed by

$$C_0 = \int_0^t (dt/10^{(T^*-T)/z}) \quad (1)$$

where C_0 is expressed as seconds at the reference temperature, t is the time in seconds, T^* is the reference temperature (°C), T is the actual temperature (°C), and z, as defined above, refers to the selected reaction. C_0 of a given heat treatment is the time required to get, at the reference temperature, the same chemical effect of the heat treatment, with reference to a well-defined reaction. In our case C_0 is expressed as seconds at 140 °C. Treatments differing in temperature and holding time can be therefore compared on the basis of heat damage, provided that the constancy of z over a limited temperature range only is taken into account.

Andrews (1985) found a relevant relationship between C_0 and the amount of lactulose in sterilized milk. He concluded that, if the time–temperature profile of a plant is known, the concentration of lactulose in treated milk can be predicted. If the plant works properly, the measured content of lactulose must be equal to the calculated value. Andrews (1984) and Resmini (1989) also suggested the use of lactulose as a criterion for distinguishing between pasteurized milk, in-container-sterilized milk, and UHT-treated milk. When a model solution in which a time–temperature-dependent reaction occurs is treated in a plant, the relationship between such reaction and C_0, or any other parameter of heat damage in milk, allows the prediction of the heat damage produced by that plant, even if its exact time–temperature profile is unknown. In a preliminary study (Coxe, 1989) carried out in our laboratory, a solution of lactose (5% w/v) at pH 8 was used as a model solution. As a result of the heat treatment, lactose turned into lactulose, which is easily detectable by HPLC. Adams et al. (1984) suggested the use of a sucrose solution to study the time–temperature relationship in a UHT indirect heat exchanger. The

† Research work supported by the National Research Council of Italy, Special Project RAISA, Sub-Project 4, Paper 1275.

0021-8561/94/1442-0360$04.50/0

method was based on sucrose inversion at an acid pH. The authors used a sucrose solution acidified to pH 2.5 with sulfuric acid to study a sterilization plant operating in the temperature range 127–149 °C. In these conditions, the inversion reaction exhibited an E_a of 25.4 ± 0.6 kcal/mol (106.3 ± 2.5 kJ/mol).

The purpose of this study was to find the relationship between heat damage in milk and the percentage of sucrose inversion in a pH 3 solution, both milk and model solution being treated following the same time–temperature profiles in a laboratory heat exchanger especially assembled for this purpose.

The investigation was based on the experimental determination of the time–temperature profiles in the plant, the kinetics of sucrose inversion in the model solution, and the reaction kinetics of some indices of heat damage in milk, such as the formation of lactulose and the loss of α-LA.

MATERIALS AND METHODS

Milk. Pasteurized homogenized whole milk was the test product for the experiment. It was industrially processed bulk milk containing more than 15.5% soluble whey protein referred to total protein. For each experiment, small lots were acquired on the market. Analyses were carried out immediately after the sterilization treatment except for lactulose, which was assayed on samples of processed milk after lyophilization at 20 °C and storage at −25 °C until the moment they were analyzed; all lyophilized samples underwent analysis at the same time.

Model Solution. A 10% sucrose solution (w/v, in distilled water), adjusted to pH 3 with concentrated sulfuric acid, was used in the experiments. A new batch of this solution was prepared for each experiment and stored at 5 °C for no longer than 24 h.

Laboratory-Scale Plant. A tubular heat exchanger (internal diameter, 2.16 mm) was employed for the heat treatment of both milk and the model solution. The plant was purposely designed so that pipes of different lengths could be assembled to vary the holding time of the process.

Silicon oil, heated by two thermostats and kept properly agitated by three mixers, was employed as a heating fluid.

The plant was provided with thermocouples to determine the time–temperature profile at any operative condition and with pressure gauges and needle valve to check and adjust pressure.

The fluid, either milk or the model solution, circulated in the plant by means of a positive pump, at a flow rate of 500 mL/h.

Evaluation of the Time–Temperature Profiles. The heating and cooling profiles were obtained by dipping sections of different lengths of a pipe in the heating or cooling fluids and by measuring the temperatures at the exit of the pipe. These measurements were performed by pumping distilled water in the pipe.

The equations describing the curves were obtained by mathematical interpolation of the experimental data.

Heat Treatments. Milk samples and the model solution were treated at temperatures of 120, 125, 130, 135, and 140 °C, with holding times of 3.1 s (140 °C) to 499.5 s (120 °C), according to the temperature (Table 1). The equivalent times, also shown in Table 1, were calculated from the experimental time–temperature profiles on the basis of eq 1, using the z value calculated for lactulose in the range 120–140 °C and, as the reference temperature, the temperature of the holding section.

C_0 Value. The C_0 value was calculated from eq 1, where the reference temperature T^* was set to 140 °C and z changed according to the reaction. z is related to E_a, which can be calculated from the reaction kinetics through the Arrhenius plot (Kessler, 1981). A computer integration program calculated the C_0 value in each treatment as the sum of three values corresponding to the heat damage produced in the heating, holding, and cooling phases, respectively.

Osmolality. The measurement of osmolality, which is directly correlated with the freezing point depression, was used to determine the sucrose inversion percentage. To correlate the sucrose inversion percentage with the cryoscope response, a calibration curve was produced from a series of 11 sucrose, glucose, and fructose solutions (in different proportions) adjusted to pH 3 with sulfuric acid. These solutions ranged from 10% sucrose to a solution containing a mixture of glucose and fructose (5% each), the latter standing for the completely inverted sucrose solution. A calibration straight line (OSM = 0.66 − 0.003[NIS], where [NIS] stands for noninverted sucrose percentage and OSM for osmolality expressed as Osmol/kg), with r^2 = 0.999, was obtained. The measurements of osmolality were carried out through an Osmomat 030 (Gonotec, Berlin, Germany); the analysis was performed on 50 μL of the sample. Results represent the mean of five replicates on each sample.

Table 1. Holding Times (t) Used for the Indirect Heat Treatments at the Holding Temperatures (T)

T (°C)	t (s)					
120	186.5 (195.7)[a]	238.5 (247.7)	290.5 (299.0)	365.0 (374.2)	447.2 (456.4)	499.5 (508.7)
125	66.6 (75.7)	76.0 (85.1)	85.2 (94.3)	103.0 (112.1)	125.0 (134.1)	134.2 (143.3)
130	18.8 (28.2)	25.1 (34.6)	30.9 (40.4)	37.8 (47.3)	54.0 (63.4)	60.3 (69.8)
135	8.9 (18.2)	15.8 (25.1)	18.8 (28.0)	25.1 (34.4)	30.9 (40.2)	37.8 (47.1)
140	3.1 (13.2)	4.7 (14.8)	8.9 (19.0)	18.8 (28.8)	25.1 (35.2)	30.9 (41.0)

[a] The corresponding equivalent times are given in parentheses (see text for the definition of equivalent time).

Whey Protein HPLC Analysis. The measurements were carried out according to the method described by Resmini et al. (1989). The method was slightly modified as follows: after the precipitation and centrifugation of casein at pH 4.6, part of the supernatant was diluted with a pH 6.6 phosphate buffer; dilution was 1:10 for pasteurized milk and 1:5 for sterilized milk samples, respectively. After filtration through a 0.22-μm Millipore filter, 20 μL of the sample underwent chromatography. Operative conditions of the HPLC analysis of whey protein were as follows: a reversed-phase PLRP-S column (150 × 4.6 mm, Polymer Laboratories Ltd., Churchstretton, U.K.); column temperature, 40 °C; a 821-FP fluorimetric detector (JASCO, Hachioji City, Japan) set at 280 (excitation) and 340 nm (emission); mobile phase (A) 0.1% trifluoroacetic acid water solution, (B) 0.1% trifluoroacetic acid acetonitrile solution; flow rate, 1 mL/min. The elution gradient, expressed as proportion of eluent B, was as follows: initial condition, 35% for 1 min; from 35 to 38% in 7 min; from 38 to 42% in 9 min; from 42 to 46% in 8 min; from 46 to 100% in 0.5 min; 100% for 0.5 min; from 100% to 35% in 1.5 min; 35% for 13.5 min. This study reports the values of α-LA only, which are expressed as the percentage of nondenatured protein ([NDα-LA]) remaining in milk after the heat treatments.

Lactulose. The measurements of lactulose were performed by HPLC according to the method described by Van Riel and Olieman (1986); the Brons and Olieman (1983) procedure, modified as follows, was adopted for sample clarification: 0.5 g of lyophilized milk was transferred into a 25-mL graduated flask; 4 mL of deionized water and 15 mL of pH 4.6 acetate buffer were added; after complete dissolution, the mixture was brought up to the mark with acetonitrile. After rest, the mixture was filtered, first through a Whatman No. 5 filter paper and then with a GV Millipore membrane (0.22 μm).

Chromatography conditions were as follows: injection volume, 100 μL; column, HPX-87P (Bio-Rad Chemical Division, Richmond, CA); column temperature, 85 °C; precolumn, deashing system (Bio-Rad); mobile phase, HPLC grade water (Merck, Darmstadt, Germany); detector, refractive index 1037A (Hewlett-Packard, Rockville, MD); detector temperature, 50 °C; flow rate, 0.6 mL/min. Lactulose content in the samples was determined through interpolation from a calibration curve (r^2 = 0.993) produced from solutions made of lactulose (Sigma, St. Louis,

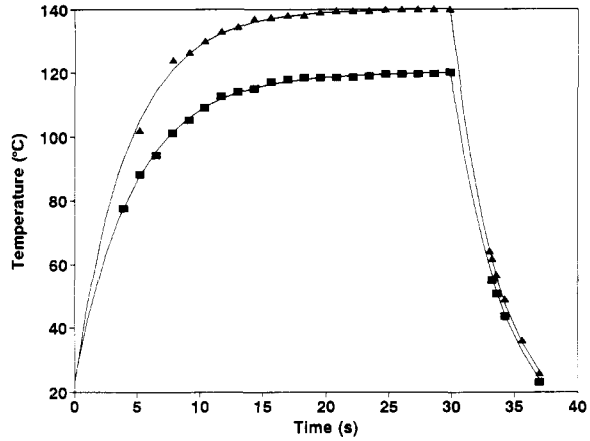

Figure 1. Time–temperature profiles in the tubular heat exchanger for the treatments at 120 (■) and 140 °C (▲).

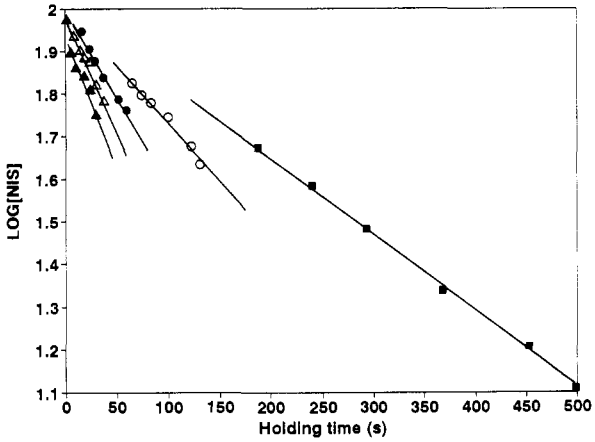

Figure 2. Sucrose inversion kinetics at 140 (▲), 135 (△), 130 (●), 125 (○), and 120 °C (■) in the model solution at pH 3. [NIS] stands for the percentage of noninverted sucrose in the model solution.

MO) (0.135–0.9 g/L) dissolved in a 50 g/L lactose solution. Results are expressed as milligrams of lactulose per gram of dry matter.

RESULTS AND DISCUSSION

Figure 1 shows the time–temperature profiles developed in the tubular heat exchanger; as an example, the treatments at 120 and 140 °C are reported.

Sucrose Inversion Kinetics. Heat treatments of the model solution (conditions in Table 1) determined the sucrose inversion kinetics represented in Figure 2, where log[NIS] was plotted against the holding times at the different temperatures. Since these relationships yielded straight lines, hydrolysis can be described by a first-order reaction. The E_a value was calculated from the sucrose inversion kinetics through the Arrhenius plot, and it was found to be 99 kJ/mol, which is close to the value of 106 kJ/mol found by Adams et al. (1984) for the inversion of a 15% sucrose solution at pH 2.5. The z value was also calculated (Kessler, 1981) and found to be 31.4 °C in the temperature range 120–140 °C. This z value was used in eq 1 to determine the value of C_0 for each heat treatment. Figure 3 shows the relation between log[NIS] and the C_0 value calculated with z = 31.4 °C. The good relation (r^2 = 0.990) found with the model solution demonstrated the accuracy with which the time–temperature profiles were determinated.

Kinetics of α-Lactalbumin Denaturation. HPLC analysis was used to determine the denaturation of the three major whey protein fractions in milk (α-lactalbumin

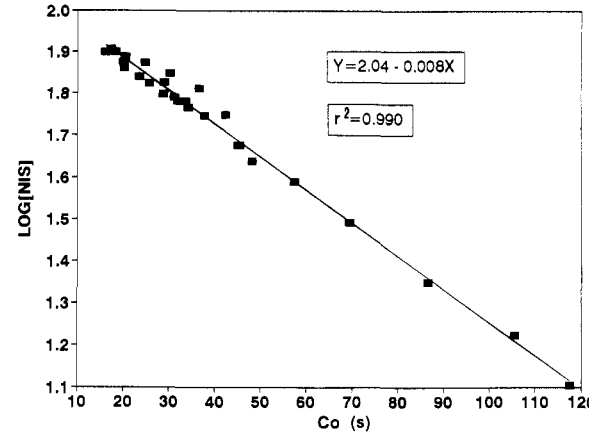

Figure 3. Correlation between the logarithm of the percentage of noninverted sucrose [NIS] in the model solution and C_0. The figure also reports the equation and the determination coefficient of the regression line.

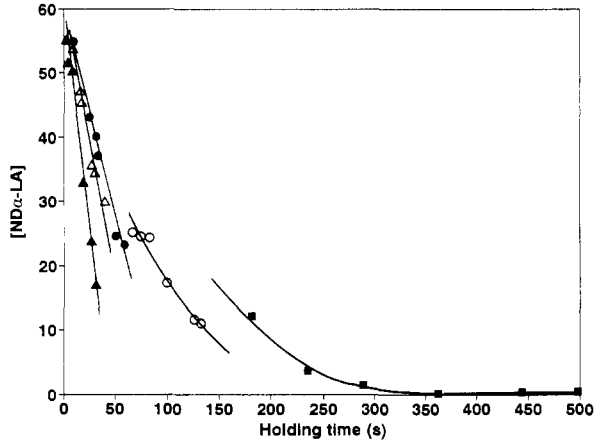

Figure 4. α-Lactalbumin denaturation kinetics in milk at 140 (▲), 135 (△), 130 (●), 125 (○), and 120 °C (■). [NDα-LA] stands for the percentage of nondenatured α-lactalbumin remaining in solution after the heat treatment of milk.

and β-lactoglobulins A and B) produced by the heat treatments. The kinetics of α-LA only are reported in this study (Figure 4), since β-lactoglobulins suffered denaturation to an extent greater than 85% as a result of the heat treatments.

α-LA denaturation was found to be a first-order reaction, with an E_a = 89.1 kJ/mol and a z value of 34.9 °C. The E_a value cannot be compared with the value found by Dannenberg and Kessler (1988), as the latter referred to skim milk and was calculated over a much wider temperature range (85–150 °C). The z value was used in eq 1 to find the value of C_0 as for α-LA. Figure 5 shows the relationship between log[NDα-LA] and the C_0 values calculated with z = 34.9 °C.

Formation of Lactulose. Lactulose formation (Figure 6) was found to follow a zero-order reaction, exhibiting an E_a = 136 kJ/mol, which is consistent with the literature results (Andrews, 1985). The calculated z value, which was found to be 22.8 °C, was used to determine C_0 relative to lactulose formation. Figure 7 shows the relationship between the amount of lactulose in milk and the values of C_0 calculated with z = 22.8 °C.

Relationships between Sucrose Inversion in the Model Solution and Heat Damage Parameters. The relations between [NIS] in the heat-treated model solution and the content of lactulose and [NDα-LA] in heat-treated milk were studied to verify the adequacy of the model solution to predict the heat damage. Linear correlations

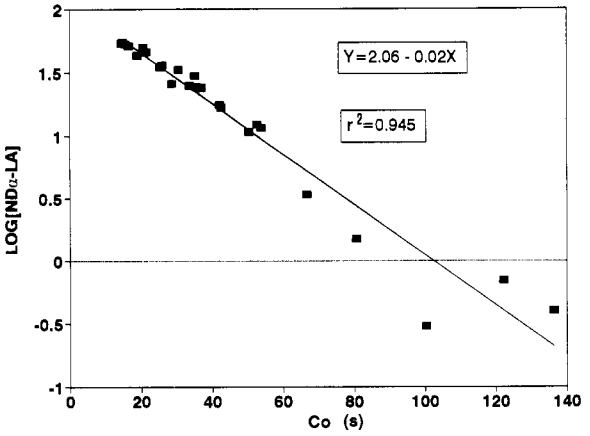

Figure 5. Correlation between the logarithm of [NDα-LA] and C_0. [NDα-LA] stands for the percentage of nondenatured α-lactalbumin remaining in solution after the heat treatment of milk. The figure also reports the equation and the determination coefficient of the regression line.

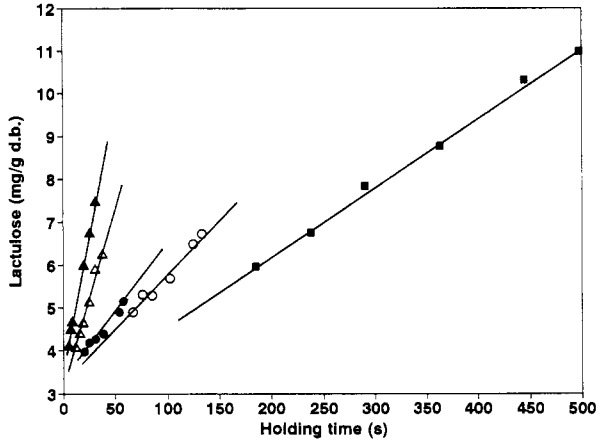

Figure 6. Lactulose formation kinetics in milk at 140 (▲), 135 (△), 130 (●), 125 (○), and 120 °C (■). Lactulose concentration is expressed as milligrams per gram of milk on a dry basis.

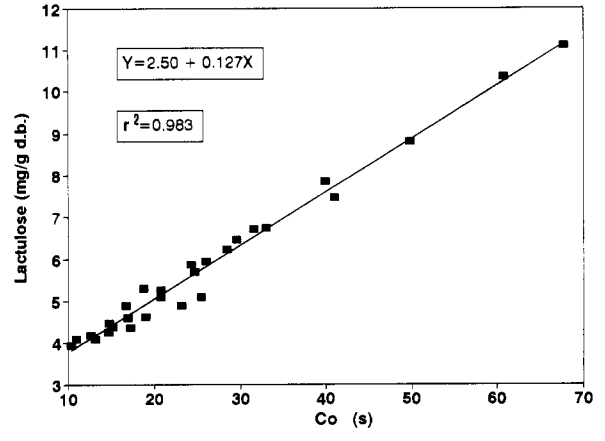

Figure 7. Correlation between the amount of lactulose formed in milk and C_0. Lactulose concentration is expressed as milligrams per gram of milk on a dry basis. The figure also reports the equation and the determination coefficient of the regression line.

(r^2 = 0.94) were found between both log[NDα-LA] and lactulose ([LACT]) in milk vs log[NIS] in the model solution (log[NDα-LA] = −4.55 + 3.30 log[NIS]; [LACT] = 20.40 − 8.43 log[NIS]).

The holding times and the equivalent times, calculated for each temperature as described under Materials and Method, are reported in Table 1.

According to Kessler (1988), the UHT treatment region

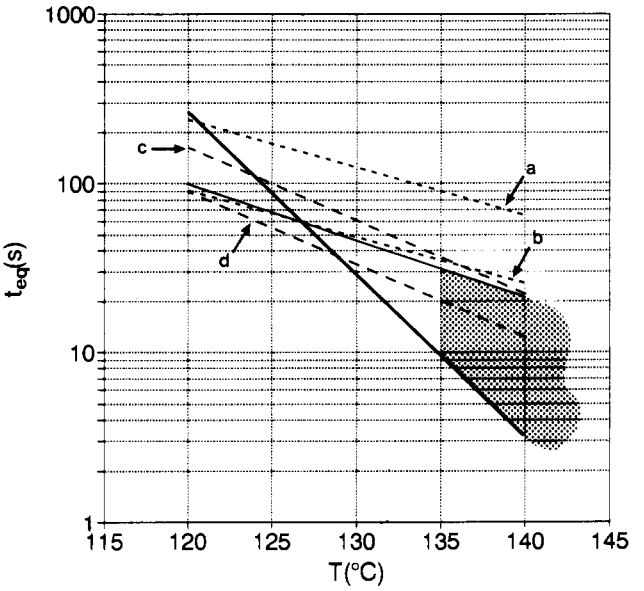

Figure 8. Diagram of the sterilization curve (heavy solid) expressed in terms of equivalent time (t_{eq}) vs temperature (T). The shaded zone between the sterilization curve and the 3% thiamin destruction curve (solid) represents the UHT region, as defined by Kessler (1988). The other curves are relative to the formation of 50 (d, broken) and 70 mg/100 mL (c, broken) lactulose, respectively, and to a remaining content of 45 (b, dotted) and 5 mg/100 mL (a, dotted) α-LA, respectively, in heat-treated milk.

is defined by the area between the curve of sterilization and the curve of 3% thiamin destruction (Figure 8). As a result of investigations on UHT-treated commercial milks, the contents of lactulose and α-LA in indirect UHT-treated milks were suggested as a means to distinguish between indirect UHT treatment and in-container sterilization. With regard to lactulose, Resmini (1989) suggested for this purpose the value of 70 mg/100 mL, Andrews found 71.5 mg/100 mL (1984), and Corzo et al. (1986) found up to 90 mg/100 mL in UHT milks, but they did not set any discriminating value for the two kinds of milk. As for α-LA, Resmini (1989) found a value of 5–45 mg/100 mL in indirect UHT-treated milks commercialized in Italy and fixed at 5 mg/100 mL the limit to distinguish them from sterilized milks.

Figure 8 reports the curves calculated for 5 and 45 mg/100 mL α-LA in milk, respectively; the former lies quite above the 3% thiamin destruction curve, while the latter (corresponding to a 60% α-LA denaturation) is close to the upper limit of the UHT region described by Kessler. Dannenberg and Kessler (1988) found the 60% denaturation curve for α-LA in the same area. On the basis of the values obtained by Resmini for this parameter, all indirect UHT-treated milks lie, in Figure 8, above the UHT treatment area described by Kessler.

On the contrary, the curve calculated for the formation of 70 mg/100 mL of lactulose lies close to the 3% thiamin destruction curve and crosses it between 140 and 145 °C. The 70 mg/100 mL lactulose curve is the upper limit of an area which is only slightly wider than the UHT treatment region suggested by Kessler. Consequently, lactulose is the most adequate parameter to check the conditions of an indirect UHT treatment of milk.

Actually, the α-LA content in commercial UHT-treated milks heavily depends also on the heat treatments that milk underwent before sterilization (pasteurization, preholding at about 90 °C, homogenization). For this reason α-LA is not a sound parameter of sterilization conditions. On the contrary, lactulose formation depends on steril-

ization conditions only, since pretreatments of milk do not produce detectable quantities of lactulose (Geier and Klostermeier, 1983; Andrews, 1984). As the effect of exposure to heat in the model solution is markedly correlated with heat damage in milk, the model solution can be used to check both the chemical and the sterilizing performances of an UHT indirect heat exchanger. A maximum and a minimum value of [NIS] in the model solution can in fact be calculated at any given sterilization temperature, to both ensure the milk sterilization and to provide a previously fixed level of quality in terms of heat damage. The following equations are employed to accomplish the above purpose: eq 2 defines the sterilization curve in the range 120–140 °C

$$\log t_{eq} = 14.0912 - 0.0971T \quad (2)$$

where t_{eq} is equivalent time (s) and T is temperature (°C), and eq 3 defines sucrose inversion at the different temperatures

$$\log[\text{NIS}] = 2.0355 + 0.0272t_{eq} - (2.4 \times 10^{-4})Tt_{eq} \quad (3)$$

where [NIS] represents the percentage of noninverted sucrose in the model solution. Equation 2 was calculated from the sterilization curve reported by Kessler (1988) using a z value of 10.3 °C as a mean value for the destruction of the thermophilic spores (Kessler, 1981); eq 3 was calculated from the data of the present study.

When checking a plant that works at a given sterilization temperature (T) (temperature in the holding section), the equivalent time (t_{eq}) is found from eq 2; then the two values (T, t_{eq}) are used in eq 3, whose result is the percentage of noninverted sucrose that ensures the commercial sterility of milk at that temperature. Higher [NIS] values correspond to heat treatments that do not guarantee the sterility of milk, at that holding temperature. Therefore, the [NIS] value obtained from eq 3 represents the maximum value that guarantees sterility at a given sterilization temperature.

The minimum percentage of noninverted sucrose required to obtain a previously stated quality in milk is calculated in the same way. For example, to get a maximum lactulose content of 50 mg/100 mL, in a continuous process working at holding temperature T, t'_{eq} is found from eq 4, defining the formation of 50 mg/100 mL of lactulose according to the kinetics reported in Figure 6:

$$\log t'_{eq} = 7.2221 - 0.04386T \quad (4)$$

Then the two values t'_{eq} and T are used in eq 3.

The following list shows the minimum and maximum [NIS] value in the model solution corresponding to a treatment producing a sterilized milk with no more than 50 mg/100 mL lactulose, at three different sterilization temperatures.

130 °C	80.0 < [NIS] < 82.8
135 °C	85.4 < [NIS] < 96.7
140 °C	90.8 < [NIS] < 100

Therefore, to produce a high-quality milk (for example, at 135 °C and assuming the limit of 50 mg/100 mL for lactulose content) in a UHT sterilization plant, a [NIS] value of 85.4–96.7% should be found when a pH 3 model solution is used. If [NIS] obtained at 135 °C is not within these values, the operative parameters of the plant should be modified until the value of sucrose falls within the above range.

The commercial sterility of milk is ensured when the model solution at pH 3 used in the tests is treated at temperatures lower than 140 °C, since at 140 °C or higher temperatures, milk sterilization could be obtained without the occurrence of sucrose inversion. This is true, for instance, at 140 °C for equivalent times lower than 5.55 s. For temperatures equal to or higher than 140 °C, the use of a model solution at a pH lower than 3 is more advisable.

Conclusions. The use of a model solution allows standardization of the process conditions of different indirect heat exchange UHT sterilization plants, regardless of their design criteria and the nominal holding times of treatments. Furthermore, the model solution is a valuable means to evaluate the effect of variations performed on both the geometrical and the operative conditions of the plant.

ABBREVIATIONS USED

α-LA, α-lactalbumin; C_0, defined for a given heat treatment as the time required to get, at the reference temperature, the same chemical effect of the heat treatment, with reference to a well-defined reaction (in our case, C_0 is expressed as seconds at 140 °C); db, dry basis; E_a, activation energy (kJ/mol); HPLC, high-performance liquid chromatography; HTST, high temperature short time; [LACT], lactulose concentration (mg/g db); [NDα-LA], percentage of nondenatured α-lactalbumin calculated as [(α-lactalbumin content in UHT treated milk)/(α-lactalbumin content in pasteurized milk)] × 100; [NIS], percentage of noninverted sucrose calculated as [(sucrose content in UHT-treated model solution)/(sucrose content in the model solution) × 100]; OSM, osmolality expressed as Osmol/kg; r^2, determination coefficient; t, time (s); T, temperature (°C); T*, reference temperature (°C); t_{eq}, t'_{eq}, equivalent time (s); UHT, ultrahigh temperature; z, increase in temperature that causes a 10-fold increase in the reaction rate (°C).

ACKNOWLEDGMENT

We express our gratefulness to Dr. Laura Pessola for collaboration in the experimental work.

LITERATURE CITED

Adams, J. P.; Simunovic, J.; Smith, K. L. Temperature histories in a UHT indirect heat exchanger. *J. Food Sci.* **1984**, *49*, 273–277.

Andrews, G. R. Distinguishing pasteurized, UHT and sterilized milks by their lactulose content. *J. Soc. Dairy Technol.* **1984**, *37*, 92–95.

Andrews, G. R. Determining the energy of activation for the formation of lactulose in heated milks. *J. Dairy Res.* **1985**, *52*, 275–280.

Brons, C.; Olieman, C. Study of HPLC separation of reducing sugars, applied to the determination of lactose in milk. *J. Chromatogr.* **1983**, *259*, 79–86.

Burton, H. Reviews of the progress of Dairy Science: The bacteriological, chemical, biochemical and physical changes that occur in milk at temperatures of 100–150 °C. *J. Dairy Res.* **1984**, *51*, 341–363.

Corzo, N.; Olano, A.; Martínez-Castro, I. Differentiation among milks processed under different thermal treatments by gas chromatographic analysis of the free disaccharide composition. *Rev. Agroquím. Tecnol. Aliment.* **1986**, *26*, 565–570.

Coxe, C. Ph.D. Thesis, University of Milan, 1989.
Dannenberg, F.; Kessler, H. G. Reaction kinetics of the denaturation of whey proteins in milk. *J. Food Sci.* **1988**, *53*, 258–263.
Erbersdobler, H. F.; Dehn, B.; Nangpal, A.; Reuter, H. Determination of furosine in heated milk as a measure of heat intensity during processing. *J. Dairy Res.* **1987**, *54*, 147–151.
Fink, R.; Kessler, H. G. HMF values in heat treated and stored milk. *Milchwissenschaft* **1986**, *41*, 638–641.
Geier, H.; Klostermeyer, H. Formation of lactulose during heat treatment of milk. *Milchwissenschaft* **1983**, *38*, 475–477.
Keeney, M.; Bassette, R. Detection of intermediate compounds in the early stages of browning reaction in milk products. *J. Dairy Sci.* **1959**, *42*, 945–960.
Kessler, H. G. *Food Engineering and Dairy Technology*; Verlag Kessler: Freising, Germany, 1981.
Kessler, H. G. Heat treatment technology of milk. *Ind. Aliment.* **1988**, *27*, 15–18.
Kessler, H. G.; Fink, R. Changes in heated and stored milk with an interpretation by reaction kinetics. *J. Food Sci.* **1986**, *51*, 1105–1111.
Mauri, L. M.; Alzamora, S. M.; Chirife, J.; Tomio, M. J. Review: Kinetic parameters for thiamine degradation in foods and model solutions of high water activity. *Int. J. Food Sci. Technol.* **1989**, *24*, 1–9.
Resmini, P. Whey proteins as indices of heat damage in milk. *Latte* **1989**, *14*, 849–853.
Resmini, P.; Pellegrino, L.; Hogenboom, J. A.; Andreini, R. Thermal denaturation of whey protein in pasteurized milk. Fast evaluation by HPLC. *Ital. J. Food Sci.* **1989**, *3*, 51–62.
Van Riel, J. A. M.; Olieman, C. High Performance Liquid Chromatography of sugars on a mixed catio-exchange resin column. *J. Chromatogr.* **1986**, *362*, 235–242.

Received for review April 16, 1993. Accepted November 22, 1993.[⊗]

[⊗] Abstract published in *Advance ACS Abstracts*, January 1, 1994.