SILLIKER LABORATORIES
Research and Laboratory Services

VALIDATION OF GROUND BEEF SHELF-LIFE EXTENSION USING ISOTHERMAL REFRIGERATED STORAGE

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Objective

The purpose of this study was to test the hypothesis that the shelf-life of ground beef stored in a refrigerator with regular defrost cycles will be less than meat stored in an isothermal (non-defrost) refrigerator.

Study Conclusion

The growth rate for spoilage bacteria under isothermal conditions was 65% of the rate for product subject to defrost cycles. This result when projected to any sample with similar characteristics predicts a 1.5-fold shelf-life improvement using 2.7-3.33ºC isothermal storage over standard refrigerated cases that defrost every 8 hours. Thus, product with a traditional shelf-life of 3.3 days would have a 5 day shelf-life when temperature is held constant.

The data from this study supports the claim that the XDX refrigeration concept can produce a significant increase in product shelf-life.

Background

A cold air refrigeration process, developed by XDX, LLC, Arlington Heights, Illinois does not require a warm air defrost cycle typical of frost-free refrigerators. Products held in an XDX modified refrigerator maintain a constant temperature.

Data was obtained from a tiered medium temperature refrigerated case in a supermarket in St. Charles, Illinois. Placement of the test probes was under the supervision of Silliker Laboratories. A data recorder was used to collect the temperatures from the probes. This data exhibited defrost cycles of 20-30 minutes every 8 hours.

Product temperatures varied with placement of thermocouples in the case. During the non-defrost cycle times, temperatures for the 3 placements average 0.2 $\pm$ 0.625°C, 3.72 $\pm$ 1.31°C, and 3.66 $\pm$ 1.56°C. During the defrost cycles product temperature increased 2.5-3.75 °C above these temperatures.

The amount of extra heat a sample is exposed to during one of the defrost cycle is represented by the area under the time-temperature curve. The average amount of heat for a cycle, obtained from the analysis of 6 cycles, was 147 $\pm$ 28 degree-minutes above background temperature.

In the laboratory, defrost cycles are simulated using a series of constant temperature water baths. Samples are placed in each bath for a defined period of time, such that the total heat input approximates field value of 147 $\pm$ 28 degree-minutes. Isothermal storage is simulated using one constant temperature water bath.

Method

TEST SAMPLES

Fresh raw aseptically ground and vacuum packaged beef maintained below 3.3°C was obtained from Vienna Beef on 1/24/00. It was repackaged in 11-gram portions in sealed waterproof plastic film pouches at the laboratory and stored at 3.3ºC until the start of the study on 1/25/00.

STORAGE CYCLES

Isothermal storage was simulated by immersing the packaged meat in a water bath. Refrigerator defrost cycles were simulated by transferring meat samples between water baths of different temperatures. Pouches were flattened to maximize the rate of heat equilibration with the bath water.

The isothermal temperature was set at 3.3°C. This temperature was obtained from data collected by UNDERWRITERS LABORATORIES who ran tests proving the temperature stability of the XDX system.

In this study, one series of samples was maintained at the isothermal temperature. A second series of samples were kept in the 3.3ºC bath between defrost cycles. Defrost cycles were scheduled every eight hours, beginning 8 hours after initiation of the study. Each cycle consisted of 4 minutes at 5.5ºC, 6 minutes at 7.2ºC, 4 minutes at 8.3ºC, 6 minutes at 7.2ºC, and 4 minutes at 5.5ºC. After the cycle samples were returned to the 3.3°C bath. The total planned added heat per cycle was thus 152 degree-minutes. Water baths were used for each of the indicated temperatures.

Thermocouples were placed in four of the test sample pouches. Two pouches were used to monitor the samples in the 3.3ºC bath and two were used to monitor temperature cycled samples.

Temperatures were recorded at 1 minute intervals using Yokagawa 3087 portable hybrid recorder (Shenandoah, GA).

SAMPLING

Test samples in triplicate were removed from constant 3.3ºC storage after 0, 24, 48, 72, 96, and 120 hours. Triplicate samples subjected to simulated defrost cycling were analyzed at the same times as the isothermal process. These samplings occurred just after cycles 3, 6, 9, 12, and 15.

ANALYSES

Samples were analyzed by plate count methods for aerobic plate count, lactic acid bacteria and coliforms. Methods of analyses are shown in the following table.

Test	Medium	Incubation Time/ Temperature/ Atmosphere
Aerobic Plate Count (APC)	Tryptose Glucose Yeast agar (TGY)	72 hours/25°C/aerobic
Lactic Acid Bacteria	DeMan, Rogasa, Sharpe (MRS) agar with overlay and cycloheximide	5 days/ 30°C/(microaerobic)
Coliforms	Violet Red Bile Agar (VRB) with overlay	24 hours/35°C/aerobic

Results and Discussion

The temperature of samples held for 5 days in the 3.3ºC water bath averaged 2.7 $\pm$ 1.75ºC. An average temperature of 2.88 $\pm$ 2.12ºC was observed for samples subjected to a simulated defrost cycle every 8 hours. The defrost cycles account for the higher average difference and standard deviation observed. The added heat of the 15 defrost cycles averaged 167 $\pm$ 58 degree-minutes.

Plate counts for the test samples are shown in Table 1. Variations in count for this type of product are expected, because the bacteria are not necessarily uniformly distributed between samples and because different bacteria grow at different rates. For the purpose of the study, the counts are examined for upward trends. Counts which appear abnormally high or low (e.g. in Table 1, the APC at 48 hours of 1,100,000 cfu/g) were discarded for this analysis.

To identify upward trends in counts, results are plotted. For this analysis, the logarithm base 10 values of the counts are used. The logarithmic transformations are necessary for standard statistical tests which require populations displaying a normal distribution. A line is drawn through the plotted points using least squares linear regression. Lines of best fit are used to determine and compare rates of growth.

Figure 1 shows the aerobic plate count results. There was little difference in growth rate. From the plot it appeared that the growth rate for the isothermal samples was faster, however, a statistical comparison of the growth rates indicated that the difference was not significant (Table 2). Low oxygen levels in the packages was the most probable reason for the low growth rates observed under either test condition.

Figure 2 shows the coliform count results. The growth rates for these organisms were lower than those observed for the aerobic bacteria (Table 2). Coliform bacteria were not expected to grow at the temperatures experienced in this study. This result demonstrates the absence of high temperature abuse.

Figure 3 shows the results for the lactic acid bacteria counts. These organisms grow under low oxygen conditions and are common spoilage organisms of packaged ground beef. The growth rates under both storage conditions were higher than for the other organism types

evaluated in this study (Table 2). Furthermore the growth rate was statistically higher for samples subjected to the simulated defrost cycles than for samples maintained without defrost cycling.

Linear regressions are valuable prediction tools. With the assumption that the lines will continue, predictions can be made as to how long it will take to increase the bacterial count from any starting level to any final level. These predictions are usually used for shelf-life assessment. The taste, odor and appearance of a food (organoleptic qualities) are the ultimate criteria used by consumers to judge a food's acceptability. These qualities begin to change as the microflora in the food- bacteria, yeast, and mold- grow and metabolize available nutrients. Organoleptic changes are generally not detectable until the microbial population is high. The number of organisms required to cause spoilage varies with the food item and the type(s) of microorganisms growing it. Generally, however, the end of shelf-life is defined as 10,000,000 bacteria per gram, 100,000 yeast per gram or visible mold.

The rates of increase of the lactic acid bacteria was the fastest of the organisms examined. Thus, these counts represent the best estimators for assessing the spoilage of the test samples. From the slopes presented in Table 2, a shelf life prediction for the product can be made. Assuming 10,000,000 colony forming units per gram as the end of shelf-life and extension of the plots in Figure 3 into the future, the shelf life of samples stored at constant temperature would have been 13.8 days while the samples subjected to cycling would have lasted 8.8 days, a difference of 5 days.

Normally, raw ground meat is not displayed for more than 5 days. To predict improvements obtained by isothermal storage, the ratio of the growth rates, 0.408510/0.266059 = 1.53, is used. The difference being an extra 0.53 days of shelf-life for each day expected.

Shelf-life in Days	
Temperature Cycled Storage	**Isothermal Storage**
1	1.5
2	3.1
3	4.6
4	6.1
5	7.7

These shelf-life predictions in this study are valid for the test materials used. Similar results are expected for samples of similar composition held under identical storage conditions.

Table 1. Validation of raw ground beef shelf-life extension using refrigerated storage with and without defrost cycling.

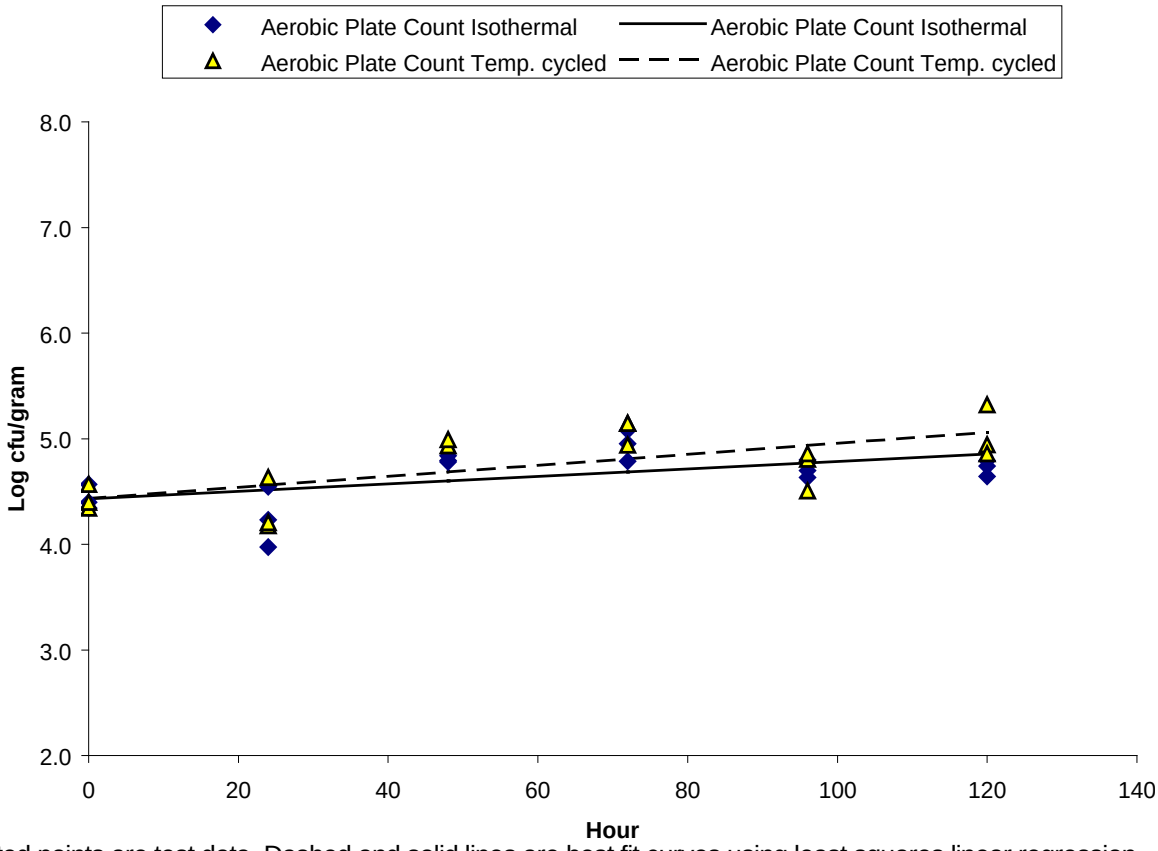
Time (hour)	Replicate	APC cfu/g		Coliforms cfu/g		Lactic acid bacteria cfu/g	
		Constant Temp.	Temp. cycled	Constant Temp.	Temp. cycled	Constant Temp.	Temp. cycled
0	1	22,000	-	6,300	-	3,200	-
	2	37,000	-	9,800	-	4,300	-
	3	25,000	-	580	-	2,400	-
24	1	35,000	15,000	5,700	2,600	2,100	940
	2	17,000	16,000	2,600	1,600	830	1,200
	3	9,400	43,000	930	3,000	1,100	1,700
48	1	62,000	1,100,000	4,500	44,000	3,400	5,800
	2	60,000	86,000	12,000	45,000	1,700	3,300
	3	69,000	98,000	10,000	29,000	1,400	3,600
72	1	90,000	87,000	7,000	32,000	270	870
	2	61,000	140,000	10,000	27,000	900	660
	3	120,000	140,000	7,200	43,000	1,300	760
96	1	50,000	64,000	58,000	130,000	63,000	3,500
	2	63,000	32,000	55,000	140,000	700	5,400
	3	43,000	72,000	42,000	99,000	3,300	1,400
120	1	62,000	88,000	74,000	220,000	3,200	3,600
	2	44,000	72,000	120,000	120,000	2,700	2,700
	3	55,000	210,000	6,400	450,000	1,300	4,700

Table 2. Linear regression analysis of plate counts.

Treatment	Growth Rate (log cfu/day)		
	Aerobic Count[a]	Coliform Count[a]	Lactic Acid Count[b]
Constant temperature	0.084762	0.018004	0.408510
Temperature cycled	0.125366	0.018341	0.266059

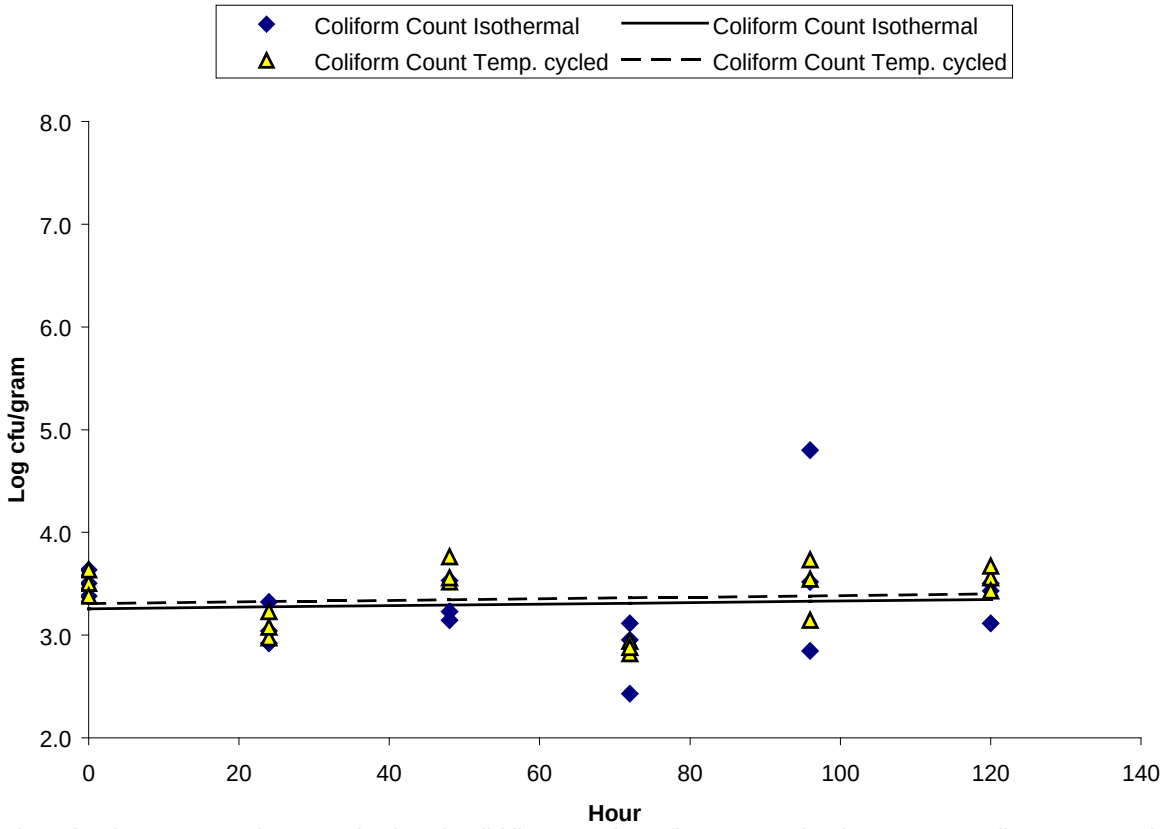
a. growth rates were not significantly different ($p>0.05$).
b. growth rates were significantly different ($p<0.05$).

Figure 1. Aerobic plate count changes of ground beef stored with and with simulated defrost cycles.



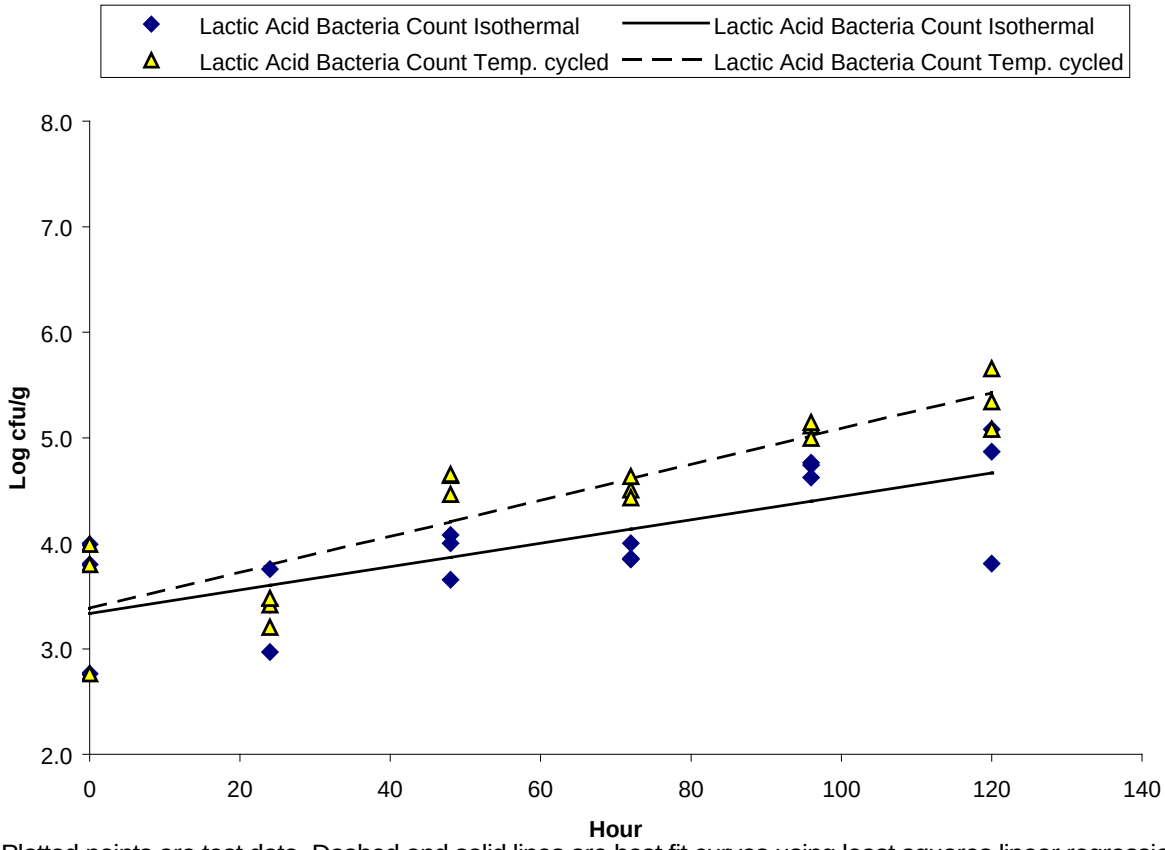
Plotted points are test data. Dashed and solid lines are best fit curves using least squares linear regression analysis.

Figure 2. Coliform count changes of ground beef stored with and with simulated defrost cycles.



Plotted points are test data. Dashed and solid lines are best fit curves using least squares linear regression analysis.

Figure 3 Lactic acid bacteria count changes of ground beef stored with and without simulated defrost cycles.



Plotted points are test data. Dashed and solid lines are best fit curves using least squares linear regression analysis.