

The Influence of Rapid Air Cooling and Carbon Dioxide Cooling and Subsequent Storage in Air and Carbon Dioxide on Shell Egg Quality^{1,2}

K. M. Keener,^{*,3} J. D. LaCrosse,^{*} P. A. Curtis,^{*} K. E. Anderson,[†] and B. E. Farkas^{*}

^{*}Department of Food Science, North Carolina State University, Raleigh, North Carolina 27695-7624; and

[†]Department of Poultry Science, North Carolina State University, Raleigh, North Carolina 27695-7608

ABSTRACT This study examined the effect of rapid cooling with air and CO₂ on shell egg quality over 14 wk. The 240 fresh eggs were initially heated to 47 C for 24 h in an incubator, cooled using rapid air cooling or CO₂ cooling, and then stored in air or CO₂ in 250-mL jars for 14 wk. The CO₂ levels were recorded of the jar atmosphere, of the egg air cell, and of the egg albumen. The Haugh units of each egg, pH, and of albumen from five eggs per group were also recorded. Haugh units are a logarithmic, empirical relationship between albumen height and egg weight (Stadelman, 1995).

Haugh units for the control eggs averaged 70.8 over 10 wk of the study. The control eggs were of such poor quality that they could not be sampled after 10 wk. The

air-cooled and CO₂-stored eggs averaged 70.3 Haugh units over the 14-wk storage period; however, the egg quality significantly deteriorated after 10 wk. The CO₂-cooled and CO₂-stored eggs averaged 75.9 Haugh units over the 14 wk study, with no observable decrease in quality.

Rapid air-cooling produces a lower quality egg than rapid cooling with CO₂. Subsequent storage of rapidly air-cooled eggs in CO₂ may increase shelf life, but Haugh units were not statistically different from rapid air-cooled eggs. CO₂-cooling and subsequent storage in CO₂ increased Haugh units. The shelf life of shell eggs could be extended to greater than 14 wk when the eggs were CO₂-cooled and CO₂-stored.

(Key words: rapid cooling, carbon dioxide, shell eggs, Haugh units)

2000 Poultry Science 79:1067–1071

INTRODUCTION

Egg production in the United States in 1997 was 182.5 million cases (at 30 dozen eggs per case). Of these, approximately 29% went to further processing, 54% went to retail, 16% went to food service, and 1.2% were exported (American Egg Board, 1998). The egg industry adds \$4 billion dollars annually to the United States. Despite the large volume of shell eggs packed for domestic use, it appears that practices and techniques used in the shell egg industry have changed little in the last 40 yr; however, the current issues of food-borne illness and microbiological contamination have brought eggs and egg processing operations to the forefront. Research has shown that *Salmonella enteritis* growth on and within eggs can be significantly retarded if eggs are stored at an internal temperature of 7 C (45 F) (Stadelman, 1995). In 1991, the United

States Congress proposed a law requiring storage and transporting of eggs to be done at or below the refrigeration temperature of 7 C (Curtis et al., 1995). On August 27, 1999, the USDA-Food Safety Inspection Service (FSIS) and Food and Drug Administration jointly enacted a refrigeration and labeling amendment to the Egg Product Inspections Act for shell eggs that states "All eggs packed for consumer use must be stored and shipped under refrigeration at an ambient temperature not to exceed 7 C (45 F)" (USDA, 1999).

Current practice in shell egg processing is to manually bring or convey eggs from the laying house into the egg packing plant. The eggs are spray-washed with a chlorinated, hot water solution (40 C) and then visually inspected and sorted. Next, the eggs are conveyed onto a computer-controlled scale, weighed, and packed into cartons based on size (e.g., small, medium, large, extra large). The cartons are packed into a case (30 dozen), and the cases are placed on a pallet (approximately 20 cases). Next, the pallet is placed in a large refrigerator to equilibrate to 7 C. The eggs in the center carton require 7 to 10 d to reach 7 C when starting at an initial temperature of 25 to 30 C (Anderson et al., 1992). Typically, eggs being sent to retail markets are shipped immediately after palleting. This shipping further increases cooling times because of the limited cooling capacity of refrigerated

Received for publication November 9, 1999.

Accepted for publication March 9, 2000.

¹Journal Paper No. FSR 99-35 of the North Carolina State University Department of Food Science Journal Series.

²This paper was presented as American Society of Agricultural Engineering (ASAE) Paper No. 99-6094 at the ASAE International Meeting in Toronto, Canada, from July 17 to 21, 1999.

³To whom correspondence should be addressed: kevin_keener@ncsu.edu.

trucks and retail outlets. Because of the elevated temperatures of the center eggs for an extended period of time, increased microbial growth occurs, and egg quality decreases (Curtis et al., 1996). Most shell egg processors put a 4-wk expiration date on the label, the limiting factor being egg quality. An increase in the shelf life of eggs also would benefit the producers interested in export opportunities.

Curtis et al. (1995) first proposed the rapid technique of cooling eggs in the carton using cryogenic gases (N_2 and CO_2). The new process reduced the cooling time from 7 to 10 d to approximately 15 to 20 min. It was proposed that using cryogenic gases to rapidly cool eggs would significantly improve egg quality and retard the growth of microorganisms.

Historically, there is a significant potential for exporting high-quality shell eggs from the United States. Approximately 96% of the world's population resides outside the United States. There is a demand for fresh shell eggs in other areas of the world where fresh eggs are not available. Carbon dioxide cooling and storage of shell eggs is a potential way of delivering high-quality, fresh eggs to these consumers.

The albumen helps position the yolk in the center of the egg and away from the shell. The quality of the albumen declines with time and storage conditions (Sharp and Powell, 1931; Hurnik et al., 1978). The exact mechanism of this albumen quality decrease with time is currently being investigated (Brake et al., 1997). The pH of a newly laid egg is between 7.6 and 8.5 (Heath, 1977); however, during storage the pH increases at a temperature-dependent rate to about pH 9.7 as CO_2 diffuses out of the egg (Sharp and Powell, 1931). It is also important to note that the buffering capacity in the egg is minimal between 7.5 and 8.5 (Cotterill and Winter, 1955). Other studies suggest that part of the albumen thinning is associated with CO_2 loss from the egg or increasing pH. Heath (1977) observed that the sulfhydryl content increases with increasing pH and suggested that this increase in sulfhydryl content and the associated albumen thinning were the result of the uncoiling of albumen proteins. Thus, temperature and storage conditions play an important part in the albumen thinning process. Wilhelm (1940) reported that albumen indices improved when eggs were stored in sealed mason jars with a CO_2 atmosphere at temperatures above -1 C. Below -1 C, no difference in albumen index was observed between eggs stored in CO_2 or in a paper carton; however, Cotterill and Gardner (1957) found that natural thinning of the albumen was not a function of temperature if pH was maintained near its initial value. The albumen index is a linear, empirical relationship between albumen height and egg weight (Stadelman, 1995). A freshly laid egg has a higher albumen index than an older egg. Thus, cooling method and storage conditions may be important in maintaining egg quality and increasing shelf life. The

objective of this study was to investigate the effect of rapid air and CO_2 cooling and storage in CO_2 or air on changes in Haugh units (quality) with age and storage conditions.

MATERIALS AND METHODS

In this study we examined the effect of rapid air and CO_2 cooling and storage on shell egg quality. An experiment was designed with three groups: air-cooled and -stored (control), air-cooled and CO_2 -stored, and CO_2 -cooled and -stored.

CO₂-Cooled Eggs

Ninety-six 2-d-old shell eggs were heated in an incubator at 47 C for 24 h. This heating was to produce eggs with temperatures comparable to those found in conventional egg washing and processing facilities. The eggs were cooled in an insulated CO_2 cooling chamber.⁴ The outside dimensions of the cooling chamber were approximately 100 × 100 × 70 cm. The cooler used liquid CO_2 under 2.07 MPa of pressure coming in through a dual manifold. The dual manifold was located approximately 15 cm above the bottom of the chamber. Uniformly spaced nozzles along the manifold delivered a fine "CO₂ snow" across the bottom of the chamber. A 43-cm diameter fan located 25 cm above the manifolds provided uniform CO_2 gas circulation throughout the chamber. The chamber temperature was maintained between -50 C and -60 C, and the chamber environment was maintained above 92% CO_2 for all cooling tests. A single rack approximately 10 cm above the fan held four flats (30 eggs per flat) of shell eggs. The four flats were symmetrically placed within the chamber with 5 cm of spacing around each flat. Preliminary laboratory studies indicated that this spacing provided uniform cooling of the eggs to 11 C for the center egg temperature in approximately 7 min. These eggs, after a 15-min equilibration in room-temperature (20 C) CO_2 , had an average temperature of 7 C.

After cooling, individual eggs were placed in 250-mL glass jars. An O_2 sensor was placed in the bottom of each jar to measure O_2 concentration as the jar was filled with ultra-high purity grade (99.9999%) CO_2 . When the O_2 level in the jar was less than 1.0% O_2 , the jar was sealed with a screw-type lid. The lids, equipped with rubber septa for syringe sampling, had permanent rubber gaskets on the inside to produce an airtight seal with the jars. The jars were then refrigerated at 7 C.

Air-Cooled and Control Eggs

One hundred forty-four eggs were heated in the same manner as the CO_2 -cooled eggs. The eggs, on six flats, were placed in a walk-in freezer at -20 C. The flats were placed in two columns in front of a 53-cm rectangular box fan. The box fan circulated cold air over the eggs for 14 min. The eggs were then removed, and 96 were sealed in CO_2 -filled jars in the same manner as the CO_2 -cooled

⁴Praxair, Inc., Burr Ridge, IL 60521.

eggs. The remaining 48 eggs were sealed in room-temperature air-filled jars for the control group. The jars were then refrigerated at 7 C. Preliminary laboratory studies indicated that after 14 min of air-cooling and 15 min of equilibration the eggs had an average temperature of 7 C.

Gas Sampling

A Fisher Gas Partition system (model 1200⁵) with two columns and a thermal conductivity detector was used for gas analyses. The first column, a ColumPak PQ,⁵ was used for CO₂ gas separation, and a second 13X molecular sieve was used for further separation of other gases (N₂, O₂, CO, H₂, and CH₄). The gas analyzer oven was operated at 51 C with ultra-high purity (99.999998%) helium carrier gas at a flow rate of 9.0 ml/min. The oven temperature and gas flow rate were optimized for maximum peak separation, and gas component percentages were determined based on ratios of total areas under the curves. We calibrated the system at initial setup, and a verification was performed every 2 wk with a calibration gas mixture⁶ of 71.84% N₂, 15.00% CO₂, 8.12% CO, and 5.04% O₂.

A Fisher GP1200 septum⁵ was glued on the blunt end of the egg (location of air cell) to form a gas-tight seal during sampling. A 1.0-ml samplelock gas-tight syringe⁷ with a 22-ga point style 3 (blunt) needle was purged with ultra-high purity helium and inserted into the air cell; 0.5 ml of gas was removed and injected into the gas partition system. The location of the air cell was determined using a candling light.

Haugh Units

Each egg was weighed and broken onto a breakout table. A semi-automated Haugh unit analyzer⁸ was used to measure albumen height and to calculate Haugh units.

pH

The albumen was separated from the yolk, and the yolk was discarded. The albumen was placed in an 8-ounce plastic Whirl-pak[®] bag. The albumen was mixed by hand, and the pH was measured using a pH meter (model 250A⁹) and low-maintenance pH triode. Because pH was very consistent, it was only measured on the first five eggs of each group per set of samples.

CO₂ Analysis

The CO₂ analysis method was developed from a previously published method used in pickle fermentation (Fleming et al., 1974). A patent disclosure is currently pending on its use for determining CO₂ levels in food

products with high fat or high protein content (Keener and Lacrosse, 1999). A typical large, fresh shell egg (60 g) has approximately 35 g of albumen and approximately 35 mg CO₂ (0.10% CO₂) in the albumen. For a 1.40-g sample of albumen (1.40 mg of CO₂), the standard deviation in this technique is 0.02 mg CO₂. The procedure used for determination of CO₂ level in egg albumen was as follows: 3 to 5 g of albumen was placed in a clean 250-ml glass jar. The exact mass was recorded. Next, a small open vial containing 5.0 mL NaOH was placed in the jar with the albumen. The jar was sealed with a metal screw-type lid. The lid provided an air-tight seal and also contained a rubber septum. Ten milliliters of a low pH (<1.0) acid phosphate solution was injected onto the albumen with a syringe through the septum in the lid. The jar was heated at 40 C for 24 h. During this time, all of the CO₂ in the albumen reacted with the NaOH in the vial. The vial was removed and titrated with HCl to determine the amount of CO₂ that was in the albumen.

Statistical Analyses

The experimental design was a 2 × 8 × 12 factorial for CO₂-stored eggs (air-cooled and CO₂-cooled), and a 1 × 8 × 6 factorial was used for air-cooled and air-stored eggs for treatment, week, and sample for a total of 240 samples. Data were analyzed using the two-way ANOVA option of the general linear models (GLM) procedure of SAS (SAS, 1996). The main effects and interactions were tested using residual error. Main effects were tested using appropriate test error based on significance of interaction. Least squares means, pooled standard errors, and mean standard errors were determined for all data. All statistically significant comparisons were at *P* < 0.05.

RESULTS AND DISCUSSION

The least squares mean and pooled least squares standard error are shown in Table 1 for the CO₂ level (in the jar, air cell, and albumen), Haugh Units, and pH for each treatment. There was a significant interaction between

TABLE 1. Summary of CO₂ level, Haugh units, and pH for each treatment¹

Treatment	CO ₂ ² (mg/mg)	Haugh units	pH ³
Air-cooled ⁴	0.3934 ^a ±0.0068	70.3 ^a ±0.78	6.75 ^a ±0.028
CO ₂ -cooled ⁵	0.3707 ^b ±0.0066	76.9 ^b ±0.69	6.70 ^a ±0.028
Control ⁶	0.0005 ^c ±0.0004	70.8 ^a ±1.0	9.23 ^b ±0.028

^{a-c}Means within column with no common superscript difference (*P* < 0.05).

¹Least squares mean and pooled least squares standard error are indicated for each treatment.

²Significant interaction between CO₂ level, method of treatment, and week of storage (*P* < 0.0001).

³pH was measured on a five-sample subset of each treatment for each testing week.

⁴n = 12 samples.

⁵n = 12 samples.

⁶n = 6 samples.

⁵Fisher Scientific Co., Springfield, NJ 07081.

⁶National Welders Supply Co. Inc., Raleigh, NC 27604.

⁷Hamilton, Co., Reno, NV 85901.

⁸Technology Services and Supplies, Dunnington, England.

⁹Orion Research, Inc., Beverly, MA 01915.

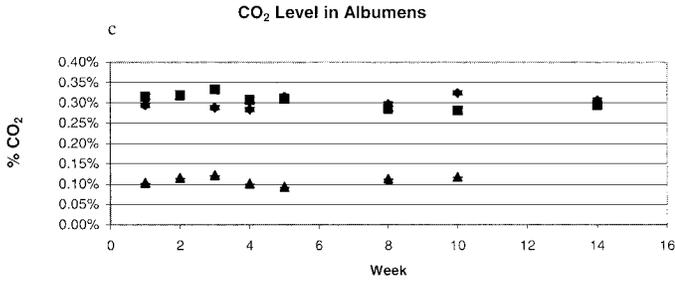


FIGURE 1. (a) Average CO₂ levels in the jar. Least squares mean and mean standard error are shown for each treatment and each week of storage. (b) Average CO₂ levels in the air cell. Least squares mean and mean standard error are shown for each treatment and each week of storage. (c) Average CO₂ levels in the albumen. Least squares mean and mean standard error are shown for each treatment and each week of storage. ◆ = air-cooled, CO₂-stored eggs; ■ = CO₂-cooled, CO₂-stored eggs; and ▲ = control eggs.

the CO₂ component (jar, air cell, and albumen), treatment, and storage week. These effects are further presented in Figure 1. Because of the order of magnitude difference between the CO₂ levels in the treatments and control eggs, the pooled least squares standard error was not representative of the true error associated with the control eggs so the mean least squares standard error was calculated. The air-cooled and CO₂-cooled eggs were stored in sealed jars that contained between 60 and 75% CO₂ over the entire 14 wk (Figure 1a). The control eggs were stored in open containers with approximately 0.014% CO₂ (Figure 1a). The CO₂ level in the air cell fluctuated from 22% to 65% over the storage period for those eggs stored in CO₂ (Figure 1b); however, no clear trends were present. The CO₂ level in the air cells of the control eggs was between 0.03 and 0.06% with a mean standard error of 0.003%. The CO₂ level in the albumen remained constant at approximately 0.30% in the CO₂-stored eggs and 0.10% in the air-stored eggs (Figure 1c). The significant interaction between the CO₂ component (jar, air cell, and albumen), treatment, and storage week appeared to be the result of unexplained variation in the CO₂ levels of the air cell and jars. For all treatments, the CO₂ levels in the albumen remained relatively constant over the 14-wk shelf life.

The control eggs, rapidly air-cooled, and air-stored eggs remained edible for only 10 wk. Mold growth and spoilage limited their shelf life. Shelf life refers to the time required for visual, undesirable changes to develop in the egg, such as mold. The air-cooled and CO₂-stored eggs were tested to 14 wk; however, we found that yolk breakage during Haugh unit testing became a problem after Week 10. In Week 10, only 5 of 12 eggs were successfully tested for Haugh units. In Week 14, only 7 of the 12 eggs were tested for Haugh units. The CO₂-cooled and -stored eggs were still in excellent condition at 14 wk with no signs of spoilage or breakage, and no problems were encountered in testing these eggs.

Table 1 shows results for the Haugh units. The CO₂ level in the control egg albumen was 0.10% on average over the 10-wk shelf life, and Haugh units were 70.8 with

a mean standard error of 1.0. The average CO₂ level in the albumen was 0.30%, but Haugh units averaged 70.3 with a mean standard error of 0.78. There was no statistical difference between the Haugh units in the control eggs (air-cooled and air-stored) and the air-cooled and CO₂-stored eggs. For the CO₂-cooled, CO₂-stored eggs, the CO₂ level in the albumen averaged 0.31% over the 14-wk period with average Haugh units of 75.9 and a mean standard error of 0.69. The Haugh units were statistically higher in the CO₂-cooled, CO₂-stored eggs (76) compared with the air-cooled, CO₂-stored eggs (70.3) and control eggs (70.8). This finding suggests that Haugh units are increased in eggs that are CO₂-cooled and CO₂-stored.

The average pH measured were 9.20, 6.75, and 6.70 for control, air-cooled and CO₂-stored, and CO₂-cooled and CO₂-stored eggs, respectively. Although not the objective of this study, it appears that pH changes from adjusting storage environment at 7 C may contribute to shelf life, but it does not contribute to higher Haugh units.

Rapid air-cooling produces a lower quality egg than rapid cooling with CO₂. Subsequent storage of rapidly air-cooled eggs in CO₂ may increase shelf life, but Haugh units are not statistically different with rapid air-cooling. Rapid CO₂-cooling and subsequent storage in CO₂ does increase Haugh units. The shelf life of shell eggs can be extended to more than 14 wk when CO₂-cooled and CO₂-stored.

ACKNOWLEDGMENTS

We acknowledge Praxair Inc. (Burr Ridge, IL 60521) for building the CO₂ cooling chamber used in this study. This research was supported, in part, by a Faculty Research and Development Grant from North Carolina State University and from state and federal funds appropriated to the Department of Food Science at North Carolina State University, Raleigh, NC 27607.

REFERENCES

- American Egg Board, 1998. Egg Industry Fact Sheet. American Egg Board, Park Ridge, IL.
- Anderson, K. E., F. T. Jones, and P. A. Curtis, 1992. Heat Loss From Commercially Packed Eggs in Post-Processing Chillers. Commercial Egg Special Report. Vol. 1. ER-1. April 1992. North Carolina Cooperative Extension Service. North Carolina State University, Raleigh, NC.
- Brake, J., T. J. Walsh, C. E. Benton, Jr., J. N. Petite, R. Meijerhof, and G. Penalva, 1997. Egg handling and storage. *Poultry Sci.* 76:144-151.
- Cotterill, O. J., and F. A. Gardner, 1957. Retarding thick white deterioration by holding shell eggs in sealed containers. *Poultry Sci.* 36:196-206.
- Cotterill, O. J., and A. R. Winter, 1955. Egg white lysozyme: The effect of pH on the lysozyme-ovomucin interaction. *Poultry Sci.* 34:679-686.
- Curtis, P. A., K. E. Anderson, and F. T. Jones, 1995. Cryogenic gas for rapid cooling of commercially processed shell eggs before packaging. *J. Food Prot.* 58:389-394.
- Curtis, P. A., K. E. Anderson, and F. T. Jones, 1996. Designing a HACCP plan for shell egg processing plants. *Egg Ind.* 101:4, 6.

- Fleming, H. P., R. L. Thompson, and J. L. Etchells, 1974. Determination of carbon dioxide in cucumber brines. *Assoc. Off. Anal. Chem. J.* 57:130-133.
- Heath, J. L., 1977. Chemical and related changes in egg albumen during storage. *Poultry Sci.* 56:822-828.
- Hurnik, G. I., B. S. Reinhart, and J. F. Hurnik, 1978. Relationship between albumen quality and hatchability in fresh and stored eggs. *Poultry Sci.* 57:854-857.
- Keener, K. M., and J. D. LaCrosse, 1999. Simple and Accurate Method for Determination of Carbon Dioxide in Food and Biological Systems. North Carolina State University (NCSU) File No. 99-70. Department of Food Science, North Carolina State University, Raleigh, NC.
- Newbold, P, 1988. *Statistics for Business and Economics*. Prentice-Hall, Inc., Englewood Cliffs, NJ.
- SAS Institute, 1996. *SAS User's Guide: Statistics*. Version 6.12. SAS Institute Inc., Cary, NC.
- Sharp, P. F., and C. K. Powell, 1931. Increase in the pH of the White and Yolk of Hen's Eggs. *J. Ind. Eng. Chem.* 23:196-199.
- Stadelman, W. J., 1995. Quality identification of shell eggs. Pages 39-66 *in: Egg Science and Technology*. W. J. Stadelman and O. J. Cotterill, ed. Haworth Press, Inc., New York, NY.
- USDA, 1999. Egg Safety. Public Release Statement 0271.99. USDA and Food and Drug Administration joint announcement. July 1, 1999. USDA, Washington, DC.
- Wilhelm, L. A., 1940. Methods of preserving interior egg quality. *Poultry Sci.* 19:3-8.