Cheesecake: A Potential Vehicle for Salmonellosis?

Y.-Y. HAO, A. J. SCOUTEN, AND R. E. BRACKETT*

Center for Food Safety and Quality Enhancement, Department of Food Science and Technology, University of Georgia, Griffin, Georgia 30223-1797, USA

ABSTRACT

This study was conducted to investigate the potential hazard of Salmonella Enteritidis surviving during the preparation and baking of cheesecake. Batters prepared with standard- and reduced-fat ingredients were inoculated with a 5-strain cocktail of S. Enteritidis (10 and 10^6 CFU/g) and were then baked according to a typical cheesecake recipe. After baking, the cheesecakes were refrigerated overnight before the survival of S. Enteritidis was determined either by direct plating or after enrichment. Samples (approximately 25 g each) were aseptically cut from the center, mid (6.35 cm from edge), and side (2.54 cm from edge) area of each cake for microbiological analysis. Proximate compositions (fat, moisture, protein, ash, pH, and water activity) of both raw batter and final baked cheesecakes were also determined. S. Enteritidis was able to survive baking of cheesecake when batter was inoculated with a high population (10^6 CFU/g) of S. Enteritidis regardless of whether standard- or reduced-fat ingredients were used. Three of nine standard- and four of nine reduced-fat cheesecake samples contained viable S. Enteritidis. In addition, one sample contained viable S. Enteritidis population detectable by direct plating (approximately 10 CFU per g of cake). This sample was taken from the center of a standard-fat cheesecake that was inoculated with a high population (10^6 CFU/g) of S. Enteritidis. Results of this study suggest that cheesecake prepared with eggs of low microbiological quality or cheesecake improperly handled or stored could serve as a vehicle for salmonellosis.

Egg-associated salmonellosis is a substantial public health problem. Salmonellae are one of the most commonly reported causes of foodborne outbreaks in the United States. These organisms have accounted for 28% of outbreaks of known etiology and 45% of foodborne disease cases associated with outbreaks of known etiology during 1973 to 1987 (3). In 1995 alone, 45,970 cases of salmonellosis were reported to the Centers for Disease Control and Prevention (5). Based on reported cases, it is estimated that there are actually 4.6 million cases of salmonellosis a year in the United States. Since 1976, salmonellosis cases caused by Salmonella Enteritidis have dramatically increased (4). The proportion of S. Enteritidis among reported Salmonella isolates increased from an estimated 5% in 1976 to 26% in 1994. The major source of S. Enteritidis is grade A table eggs (13).

Unlike most Salmonella species, which typically contaminate only the eggshell, S. Enteritidis can be found inside eggs. Hence, normal washing practices will not be able to eliminate this bacterium from eggs. Without proper cooking, S. Enteritidis can still cause illness even if shell eggs are properly cleaned.

There are many foods that rely on raw or mildly heated eggs to attain specific texture or flavor, e.g., French silk pie, meringue, mousse, Bavarian cream, and Hollandaise sauce. Although consumers are likely to tremale or heat breakfast eggs well done, the potential hazard for foods prepared using relatively mild heating has not been well addressed. Cheesecake is a popular food that may fall into this category. Cheesecake is prepared using cream cheese and eggs as primary ingredients and is usually cooked at a relatively low temperature for a longer time. It would be reasonable to suspect that improper preparation of cheesecakes could lead to potential foodborne illness. Indeed, cheesecakes prepared with raw shell eggs were suspected in two outbreaks causing illness in 18 people in Cherwell and Oxfordshire, England, in 1996 (6, 7). The objective of this study was to confirm our hypothesis that S. Enteritidis could survive the baking process of cheesecake making.

MATERIALS AND METHODS

Salmonella Enteritidis. Five strains of S. Enteritidis, E 565-88 (tuna salad), E 180-88 (human), E 457-88 (egg), SE 61697 (human), and E 1294 (human, egg outbreak), were used in the study. All cultures were maintained on tryptic soy agar (TSA; Difco Laboratories, Detroit, Mich.) slants at 4°C. Cultures were activated in tryptic soy broth (Difco) at 37°C at least twice for 24-h periods prior to being used. Twenty-four-hour cultures of each strain were sedimented by centrifugation (approximately 1,800 × g, 10 min), (IEC Clinical Centrifuge, International Equipment Co, Needham Heights, Mass.), and pellets were resuspended in 0.1 M of potassium phosphate buffer (pH 7.0) to give a population of approximately 6.5 × 10^9 CFU/ml of buffer. A 5-strain cocktail was prepared by combining approximately equal portions of each strain and diluting in phosphate buffer to give final populations of about 10 and 10^6 CFU/g of batter.

Preparation of samples. All ingredients used in this study were purchased at a local retailer and kept in the refrigerator until used. Eggs, cream cheese, and margarine were removed from the refrigerator and allowed to stand at room temperature (approximately 30 min) to soften cream cheese and margarine as required in the recipe. Crust was prepared by combining graham cracker
crumbs (1 cup, approximately 100 g), granular sugar (3 tablespoons, approximately 46 g), and melted margarine (3 tablespoons, approximately 42 g). The mixture was pressed onto the bottom of a 9-in springform pan and baked at 163°C for 10 min in a Kenmore oven (model 911.93273790; Sears, Roebuck & Co., Ill.).

Batter was prepared with softened cream cheese (approximately 907 g), granular sugar (1 cup, approximately 100 g), and flour (3 tablespoons, approximately 35.4 g) and mixed using a mixer equipped with a whisk (speed 4; model KSM5; Kitchen Aid, St. Joseph, Mich.) until well blended. Four eggs (grade A) were added, one at a time, mixing well after each addition. At this point, 1 ml of low- or high-inoculum cocktail (approximately 10^4 or 10^9 CFU/ml, respectively) was added to the batter and mixed for 2 minutes to achieve a final inoculation level of 10 or 10^6 CFU/g of batter, respectively. Sour cream (1 cup, approximately 120 g) and vanilla extract (1 tablespoon, approximately 16 ml) were added and mixed into the batter with a spatula. The batter was poured over the crust and baked, one at a time, in the preheated Kenmore oven at 232°C for 10 min then for another hour at a reduced temperature (121°C). The cakes were removed from the oven, cooled at room temperature for approximately 2 h, then stored at 5°C overnight before sampling.

Reduced-fat cheesecakes were prepared by replacing standard-fat cream cheese and sour cream with commercially available ½-less-fat cream cheese and 30%-less-fat sour cream, respectively.

Temperatures were monitored during baking using a digital thermometer (Omega, Stamford, Conn.) with copper-constantan, T-type thermocouples. The thermocouples were located at the center, mid (6.35 cm from edge), and side (2.54 cm from edge) areas of the pan and also in the oven. Temperatures were recorded at beginning of baking, after 10 min, and every 15 min thereafter.

**Microbiological analysis.** Three samples of approximately 25 g each were aseptically cut from the center, mid, and side area of each cake. Each sample was blended with 25 ml of phosphate buffer, and 0.1 ml was deposited and evenly distributed on plates of TSA (Difco) using a bent glass rod. Plates were incubated at 37°C for 48 h. TSA plates containing visible colonies were then replica plated (10) onto XLD agar (Difco) and then incubated at 37°C for 48 h. Enrichment was conducted by transferring 10 ml of blended sample to 90 ml of lactose broth (Difco) and incubated at 37°C for 48 h. The broth was then streaked on XLD agar to detect the presence of *Salmonella*.

Presumptive *Salmonella* isolates were confirmed with the API 20E analytical profile index (Analytab Products, Division of Ayerst Laboratories Inc., Plainview, N.Y.).

**Chemical analysis.** Duplicate samples of both raw batter and cooked cakes were subjected to chemical analysis. Moisture content was determined using a vacuum-oven method (Association of Official Analytical Chemists, Procedure 955.30) (2). Crude fat content (%) of moisture-free samples was determined using a Goldfisch extractor (model 3500; Laboratory Construction Co., Kansas City, Mo.) (American Association of Cereal Chemists, method 08-01) (1). Nitrogen content was determined using an automatic nitrogen analyzer (LECO FP-2000; LECO Corp., St. Joseph, Mich.) according to instructions of the manufacturer with EDTA (LECO Corp., 9.85 ± 0.03% N) as a reference standard. A protein conversion factor of 6.25 was used to calculate the protein content (2). Ash content (%) was determined using procedure 923.03 of the Association of Official Analytical Chemists (2). Water activity was determined using an Aqua Lab CX-2 meter (Decagon Devices Corp., Pullman, Wash.).

**Statistical analysis.** Three independent replicate trials were conducted. Temperature data were analyzed using the General Linear Model procedure (P ≤ 0.05) (14).

### RESULTS AND DISCUSSION

The temperature profile of cheesecake during baking is illustrated in Figure 1. Temperature increased from an initial value of approximately 18°C (batter) to a final temperature of approximately 68°C at the center of the cake immediately before the cakes were removed from the oven. Fat content did not affect the temperature profile as expected. However, internal temperature differed by more than 10°C between the side sections and the center of the cakes.

**TABLE 1.** Proximate composition, pH, and water activity of batter and cheesecakesa

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>pH</th>
<th>Water activity</th>
<th>Moisture content (%)</th>
<th>Crude fat (%)</th>
<th>Carbohydrate (%)</th>
<th>Protein (%)</th>
<th>Ash (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Batter</td>
<td>Standard fat</td>
<td>4.96 ± 0.02a</td>
<td>0.98 ± 0.001</td>
<td>52.6 ± 0.21</td>
<td>19.4 ± 0.40</td>
<td>20.0 ± 0.00</td>
<td>6.1 ± 0.05</td>
</tr>
<tr>
<td></td>
<td>Reduced fat</td>
<td>4.95 ± 0.01</td>
<td>0.98 ± 0.001</td>
<td>59.5 ± 0.19</td>
<td>10.8 ± 0.48</td>
<td>21.1 ± 0.00</td>
<td>7.5 ± 0.07</td>
</tr>
<tr>
<td>Cheesecake</td>
<td>Standard fat</td>
<td>4.93 ± 0.02</td>
<td>0.98 ± 0.001</td>
<td>49.1 ± 0.29</td>
<td>24.5 ± 1.59</td>
<td>18.8 ± 0.00</td>
<td>6.6 ± 0.03</td>
</tr>
<tr>
<td></td>
<td>Reduced fat</td>
<td>4.98 ± 0.05</td>
<td>0.98 ± 0.002</td>
<td>55.1 ± 0.36</td>
<td>15.3 ± 0.76</td>
<td>20.0 ± 0.00</td>
<td>8.3 ± 0.17</td>
</tr>
</tbody>
</table>

a Values are mean ± SD.

b Calculated by difference.
Fat content of ingredients used to prepare the cakes affected the proximate compositions but not pH and water activity of the final products (Table 1). Cakes made of reduced-fat substitutes retained higher moisture content, carbohydrate, protein contents, and ash but much less fat (15.3% compared with 24.5% in the standard-fat cake). The two reduced-fat ingredients used, ⅔-less-fat cream cheese and 30%-less-fat sour cream, were available in retail stores. In preliminary experiments, fat-free egg substitute was also used to replace shell eggs; however, the cake prepared from egg substitute as well as other reduced-fat ingredients resulted in a cheesecake with a soft texture and undesirable appearance. Therefore, we decided not to use egg substitute in the study.

Salmonella Enteritidis was able to survive the cooking procedure for cheesecake preparation; however, the initial inoculation level significantly affected survival of this bacterium. In no case were viable Salmonella isolated from cheesecakes inoculated with low populations (10 CFU/g) of S. Enteritidis. In contrast, 7 of 18 samples inoculated with a high population (10<sup>6</sup> CFU/g) of S. Enteritidis yielded viable Salmonella after enrichment (Table 2). In addition, one sample yielded viable S. Enteritidis on direct plating, indicating viable population of approximately 10 CFU of Salmonella per g of cake.

Many factors, such as pH, water activity, and nature of constituents in food, affect heat resistance of microorganisms. Fat and proteins have a protective effect on microorganisms, whereas higher humidity and water activity usually enhance lethality (9). We included cakes made of reduced-fat ingredients to determine if lower fat content reduces the survival of S. Enteritidis. However, results indicated that fat content in the batter (cake) tested in the study did not significantly affect survival of the bacterium (P = 0.05).

Location in cakes would determine the possibilities of finding survivors of S. Enteritidis. About 17% of side-section samples contained viable S. Enteritidis, whereas approximately 50% of both center- and midsection samples yielded viable organisms. Temperature undoubtedly accounts for the difference. There was little difference in temperature between the center and midsections; however, the temperature in the side-section increased much faster during baking and was more than 10°C higher than the center and midsections at the end of cooking.

Decimal reduction times (D value) for S. Enteritidis vary depending on the temperature, heating medium, and strain tested. D<sub>60</sub>C of this bacterium could be 0.22 min (strain PT13a) or 0.44 (PT4) (10). Humphrey (10) also reported that S. Enteritidis had a D<sub>60</sub>C of 0.3 min in whole egg but increased to 0.8 min in egg yolk, which had much higher fat than whole egg. D values of the 5-strain cocktail used in the study have been calculated by heating in egg products in a previous study in our laboratory (12). In that study, D<sub>60</sub>C was 0.04 min in whole egg, 0.21 min in egg yolk + 10% salt, and 1.26 min in egg yolk + 5% salt + 5% sugar. Sugar and fat content in batter or cake could also affect the heat resistance of S. Enteritidis; however, the magnitude of the effect they would have is unknown. In addition, the nearly 9% difference in fat did not affect the survival of S. Enteritidis in cake prepared with reduced-fat ingredients. The temperature at the center and midsections was at least 62°C after baking for 55 min, and the temperature kept rising to approximately 68°C and higher at the end of baking (another 15 min). With batter inoculated with 10<sup>6</sup> CFU/g, a minimal 6-D reduction would be needed to ensure no survival of the bacterium. No data regarding D value in such products are currently available, but based on our results, it was not possible to practically achieve such heat treatment by common preparation procedure for cheesecake and still have a cheesecake of acceptable quality.

Results of this study suggest that it is possible for S. Enteritidis to survive the heating conditions encountered during the baking of cheesecake. However, one would not expect to encounter as high a population of Salmonella in batter as used in the study, especially when the cake is prepared with good-quality eggs and proper handling and preparation practices are followed. Moreover, contamination of individual eggs with S. Enteritidis is infrequent, and outbreaks are typically associated with food service situations in which eggs are pooled (8). Nevertheless, improper storage of ingredients and negligence in preparation could increase the risk of growth of Salmonella. In addition, cheesecake should be refrigerated to prevent outgrowth of any microorganisms that might survive or contaminate the cake after cooking.

ACKNOWLEDGMENTS

We thank Jennifer Christian and Cindy Michalski for assisting with sample preparation.

REFERENCES

4. Centers for Disease Control and Prevention. 1996. Outbreaks of Sal-

### TABLE 2. Survival of S. Enteritidis in cheesecake prepared from batter inoculated with a high population (10<sup>6</sup> CFU/g) of the bacterium

<table>
<thead>
<tr>
<th>Location</th>
<th>Standard fat</th>
<th>Reduced fat</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Central</td>
<td>1/3&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2/3</td>
<td>3/6 (50%)</td>
</tr>
<tr>
<td>Mid</td>
<td>2/3</td>
<td>1/3</td>
<td>3/6 (50%)</td>
</tr>
<tr>
<td>Side</td>
<td>0/3</td>
<td>1/3</td>
<td>1/6 (16.7%)</td>
</tr>
</tbody>
</table>

<sup>a</sup> Numbers in numerator represent samples positive for S. Enteritidis by enrichment procedure. Denominators are total samples tested.

<sup>b</sup> Sample showed positive result from enrichment procedure, also had detectable population of S. Enteritidis by direct platting (approximately 10 CFU of S. Enteritidis per g of cake).


