Evaluation of Consumer-Style Cooking Methods for Reduction of *Escherichia coli* O157:H7 in Ground Beef

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ABSTRACT

The objective of this study was to evaluate the thermal inactivation of *Escherichia coli* O157:H7 in ground beef cooked to an internal temperature of 71.1°C (160°F) under conditions simulating consumer-style cooking methods. To compare a double-sided grill (DSG) with a single-sided grill (SSG), two different cooking methods were used for the SSG: for the one-turnover (OT-SSG) method, a patty was turned once when the internal temperature reached 40°C, and for the multiturnover (MT-SSG) method, a patty was turned every 30 s. Patties (100 g, n = 9) inoculated with a five-strain mixture of *E. coli* O157:H7 at a concentration of 10⁶ CFU/g were cooked until all three temperature readings (for two sides and the center) for a patty were 71.1°C. The surviving *E. coli* O157:H7 cells were enumerated on sorbitol MacConkey (SMAC) agar and on phenol red agar base with 1% sorbitol (SPRAB). The order of the cooking methods with regard to the cooking time required for the patty to reach 71.1°C was as follows: DSG (2.7 min) < MT-SSG (6.6 min) < OT-SSG (10.9 min). The more rapid, higher-temperature cooking method was generally more effective (P < 0.01) in destroying *E. coli* O157:H7 in ground beef. *E. coli* O157:H7 reduction levels were clearly differentiated among treatments as follows: OT-SSG (4.7 log₉ CFU/g) < MT-SSG (5.6 log₉ CFU/g) < DSG (6.9 log₉ CFU/g). Significantly larger numbers of *E. coli* O157:H7 were observed on SPRAB than on SMAC agar. To confirm the safety of ground beef cooked to 71.1°C, additional patties (100 g, n = 9) inoculated with lower concentrations of *E. coli* O157:H7 (10⁴ to 10⁵ CFU/g) were tested. The ground beef cooked by the OT-SSG method resulted in two (22%) of nine samples testing positive after enrichment, whereas no *E. coli* O157:H7 was found for samples cooked by the MT-SSG and DSG methods. Our findings suggest that consumers should be advised to either cook ground beef patties in a grill that cooks the top and the bottom of the patty at the same time or turn patties frequently (every 30 s) when cooking on a grill that cooks on only one side.

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whether ground beef is cooked by observing the color and texture of the cooked meat.

Several studies pertaining to the heat resistance of \textit{E. coli} O157:H7 in ground beef cooked by water bath and grilling methods have been documented \cite{7, 9, 11, 14, 17, 26}. However, information used to establish cooking recommendations has been generated largely from laboratory experiments that have not always simulated the actual cooking processes \cite{7}. Moreover, there are few reports concerning the destruction of \textit{E. coli} O157:H7 with consumer-style cooking methods. The objective of this study was to evaluate the thermal inactivation of \textit{E. coli} O157:H7 in ground beef patties cooked to an internal temperature of 71.1°C under conditions simulating consumer-style cooking methods. In addition, the effectiveness of phenol red agar base supplemented with 1% sorbitol medium (SPRAB) for the quantitative determination of heat-injured \textit{E. coli} O157:H7 in ground beef was compared with that of sorbitol MacConkey (SMAC) agar medium.

\section*{Materials and Methods}

\textbf{Bacterial cultures.} Five strains of \textit{E. coli} O157:H7 (ATCC 35150, ATCC 43889, ATCC 43890, ATCC 43894, and ATCC 43895) were obtained from the Food Science and Human Nutrition Culture Collection at Washington State University, Pullman, Wash. All cultures were maintained on tryptic soy agar (Difco Laboratories, Detroit, Mich.) slants at 4°C and subcultured monthly.

\textbf{Cell suspension.} Each of the five \textit{E. coli} O157:H7 strains was grown separately for 18 h at 37°C in 50 ml of tryptic soy broth (Difco) supplemented with 1% glucose. Cells were harvested by centrifugation at 4,000 \times \text{g} for 20 min at 4°C and washed twice with buffered peptone water. The final pellet was resuspended in buffered peptone water to a concentration calculated to yield 10^9 CFU/ml. Next, all five strains were combined in a culture cocktail, which was used as an inoculum.

\textbf{pH measurement and inoculation.} Raw ground beef (20% fat and 80% lean) was purchased at a grocery store in Pullman, Wash. All experiments were carried out with fresh ground beef. The day the beef was ground was indicated on the packaging. The pH of the ground beef was determined in three locations per package with a spear-type combination electrode (Model 430, Corning Inc., Corning, N.Y.). pHs ranged from 5.68 to 6.08. The inoculum was added to ground beef as follows: ground beef was transferred to a sterile Hobart mixer bowl and inoculated with a five-strain \textit{E. coli} O157:H7 cocktail to yield approximately 10^7 CFU/g of sample. Inoculated ground beef was mixed for 2 min with gloved hands for uniform distribution of the microorganism.

\textbf{Ground beef patties.} Weighed portions (100 g) of ground beef were placed in a plastic hamburger mold and evenly distributed inside the mold with gloved hands. The meat was pressed and flattened with a spatula and reshaped in the patty mold to produce a patty of uniform thickness (1 cm) and diameter (9 cm). Patties were stored at 4°C until they were cooked (within 40 min). Uncooked ground beef patties inoculated with the \textit{E. coli} O157: H7 culture cocktail served as controls.

\textbf{Cooking of patties.} For consumer-style cooking, an electric griddle (preheated to 163°C, Model 07035, National Presto Industries Inc., Eau Claire, Wis.) was used as a single-sided grill (SSG), and a double-sided (clam-shell) grill (DSG, preheated for 10 min; Model GR10AWHT, Salton Inc., Mt. Prospect, Ill.) was used for double-sided grilling. The SSG was tested with two different methods, the one-turnover (OT-SSG) method and the multiturnover (MT-SSG) method. For the OT-SSG method, a patty was turned once when the internal temperature reached 40°C. For the MT-SSG method, a patty was turned at 30-s intervals. A sterile spatula was used each time a patty was turned. Three type K thermocouples (Omega Engineering Inc., Stamford, Conn.) were inserted from the side of the patty into the locations of patties along radii. All patties were cooked until all three thermocouples indicated temperatures of \(\geq 71.1°C\), and the patties were then immediately removed from the griddle. Thermocouple signals were recorded every 30 s to monitor the heat flow relative to the cooking method. Cooking times were also recorded for each patty. The patties were cooked one at a time.

\textbf{Microbial enumeration.} Cooked patties were placed in stomacher bags containing 100 ml of buffered peptone water and homogenized for 2 min with a stomacher (400 Circulator, Seward, London, UK). Aliquots (1 ml) of sample were serially diluted in 9 ml of sterile buffered peptone water. Tenfold serial dilutions were carried out, and 0.1 ml of sample or diluent was plated on SMAC agar (Difco) and phenol red agar base (Difco) with 1% sorbitol (SPRAB) in duplicate to enumerate \textit{E. coli} O157:H7 and heat-injured \textit{E. coli} O157:H7, respectively. All plates were incubated at 37°C for 24 to 48 h, and then colonies were enumerated.

\textbf{Statistical analysis.} For each cooking method, three patties were cooked per day, and the experiments were performed three times (\(n = 9\)). The average of the duplicate plate counts for three repetitions was converted to units of log_{10} CFU/g. Data were analyzed by analysis of variance with the GLM procedure of SAS (Version 8.1, SAS Institute Inc., Cary, N.C.) for a completely randomized block design. The cooking method effect was tested with block \(\times\) cooking method as an error term. When the main effect was significant (\(P < 0.05\)), mean separation was accomplished with the probability option (PDIFF, a pairwise \(t\) test).

\section*{RESULTS AND DISCUSSION}

Mean cooking times taken for patties to reach an internal temperature of 71.1°C ranged from 2.7 to 10.9 min, depending on the cooking method (Table 1). In this study, each patty was cooked until all three temperature readings for that patty were \(\geq 71.1°C\). There were significant differences (\(P < 0.01\)) among cooking methods with regard to the cooking times required to reach a temperature of 71.1°C. The order of the cooking methods with regard to required cooking time was as follows: DSG (2.3 to 3.0 min) < MT-SSG (5.5 to 7.4 min) < OT-SSG (9.0 to 12.4 min). When the temperature readings for the three sites in the patty were analyzed (Table 1), wide variation in within-patty temperatures was observed for the OT-SSG method. These temperatures ranged from 1.6 to 13.3°C, indicating...
that there is a possibility that portions of the patty could be undercooked even when the center of the patty has reached 71.1°C. The lowest temperature reading was not consistently that for the middle of the patty. These internal temperature differences could affect the survival of \( \text{E. coli} \) O157:H7 in cooked ground beef patties, because some portions of the patty never reach the target temperature, while others do. In contrast, the level of variation in cooking temperatures within patties cooked by the MT-SSG and DSG methods was much lower than that for patties cooked by the OT-SSG method. These results suggest that the MT-SSG and the DSG methods are more effective than the OT-SSG for the uniform cooking of ground beef patties.

To compare the rates of \( \text{E. coli} \) O157:H7 reduction in ground beef patties cooked to 71.1°C by the different cooking methods, a high inoculation level (\( >10^7 \text{ CFU/g} \)) was used. There were significant differences \( (P < 0.01) \) in levels of \( \text{E. coli} \) O157:H7 reduction among cooking methods (Fig. 1). The \( \text{E. coli} \) O157:H7 reduction levels were clearly differentiated among treatments \( (P < 0.05) \) as follows: OT-SSG (a reduction of 4.7 \( \log_{10} \text{ CFU/g} \)) < MT-SSG (a reduction of 5.6 \( \log_{10} \text{ CFU/g} \)) < DSG (a reduction of 6.9 \( \log_{10} \text{ CFU/g} \)). These results indicate that the DSG system is more effective in destroying \( \text{E. coli} \) O157:H7 in ground beef than the SSG system is. This finding is in agreement with results obtained by D’sa et al. (11), who compared food service ground beef patties cooked with SSG and those cooked with DSG cooking systems with regard to the reduction of microbial counts. These investigators reported that cooking with the DSG system reduced \( \text{E. coli} \) O157:H7 populations in ground beef patties by 5.7 and 6.1 \( \log_{10} \text{ CFU/g} \) while SSB cooking reduced populations by 1.3 and 2.9 \( \log_{10} \text{ CFU/g} \) when patties were cooked to target temperatures of 60 and 68°C, respectively. For the SSG system, the MT-SSG method rapidly achieved an internal target temperature (Table 1) and a larger \( \text{E. coli} \) O157:H7 reduction (Fig. 1) than the OT-SSG method did. Juneja et al. (15) reported that the heating of ground beef patties to an internal temperature end point of 68.3°C resulted in a 4-\( \log_{10} \) reduction of \( \text{E. coli} \) O157:H7. Clavero et al. (7) reported that cooking ground beef patties to an internal temperature of at least 68.3°C and holding the patties at this temperature for 5 s is sufficient to destroy 4 \( \log_{10} \) CFU of \( \text{E. coli} \) O157:

<table>
<thead>
<tr>
<th>TABLE 1. Cooking properties of ground beef patties (n = 9) cooked to 71.1°C</th>
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<tbody>
<tr>
<td>Cooking method</td>
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<tr>
<td>----------------</td>
</tr>
<tr>
<td>OT-SSG</td>
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<tr>
<td>MT-SSG</td>
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<tr>
<td>DSG</td>
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</table>

*OT-SSG, one-turnover cooking with single-sided grill; MT-SSG, multiturnover cooking with single-sided grill; DSG, double-sided (clam-shell) grill.*

H7 in the patties. These investigators also stated that the internal temperature of patties would likely remain at 68.3°C for 10 to 15 s before decreasing, thereby providing additional lethality.

Following heat treatment, sublethally injured foodborne pathogens could assume added significance because they are potentially as dangerous as their uninjured counterparts (18, 19). Clavero and Beuchat (6) demonstrated the ineffectiveness of SMAC agar for the enumeration of heat-stressed \( \text{E. coli} \) O157:H7 cells. We investigated heat-injured \( \text{E. coli} \) O157:H7 in heat-processed ground beef patties with the use of SPRAB, which can recover heat-injured \( \text{E. coli} \) O157 cells (6). Table 2 shows populations of heat-injured \( \text{E. coli} \) O157:H7 in ground beef patties cooked to 71.1°C as enumerated on SMAC agar and SPRAB. Significantly larger numbers of \( \text{E. coli} \) O157:H7 cells were detected on SPRAB than on SMAC agar for the OT-SSG method (with

<table>
<thead>
<tr>
<th>TABLE 2. Enumeration of heat-injured ( \text{E. coli} ) O157:H7 from inoculated ground beef patties cooked to 71.1°C with sorbitol MacConkey (SMAC) agar as a selective medium and with sorbitol phenol red agar base (SPRAB) as a recovery medium</th>
</tr>
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<tbody>
<tr>
<td>( \text{E. coli} ) O157:H7 population (log_{10} \text{ CFU/g}) (n = 9)*</td>
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<tr>
<td>Before cooking</td>
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<tr>
<td>SMAC agar</td>
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<td>SPRAB</td>
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<td>SEM</td>
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</table>

*OT-SSG, one-turnover cooking with single-sided grill; MT-SSG, multiturnover cooking with single-sided grill; DSG, double-sided (clam-shell) grill. Means with different letters are significantly different \( (P < 0.05) \).
TABLE 3. Survival of *E. coli* O157:H7 in ground beef patties inoculated (10^3–10^4 CFU/g) and cooked to an internal temperature of 71.1°C as determined by plating immediately and by enrichment

<table>
<thead>
<tr>
<th>Block</th>
<th>Sample</th>
<th>Initial level^b^</th>
<th><em>E. coli</em> O157:H7 survival with OT-SSG</th>
<th><em>E. coli</em> O157:H7 survival with MT-SSG</th>
<th><em>E. coli</em> O157:H7 survival with DSG</th>
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<tbody>
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<td></td>
<td></td>
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<td>Direct planting</td>
<td>Enrichment</td>
<td>Direct planting</td>
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<tr>
<td>3</td>
<td>9</td>
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</table>

^a^ OT-SSG, one-turnover cooking with single-sided grill; MT-SSG, multiturnover cooking with single-sided grill; DSG, double-sided (clam-shell) grill. –, no growth on SMAC agar, +, growth on SMAC agar.

\(^b^\) Level of *E. coli* O157:H7 inoculated into ground beef patties before cooking (log_{10} CFU/g).

A mean difference of 0.8 log_{10} CFU/g; \(P < 0.05\) and for the MT-SSG method (with a mean difference of 1.1 log_{10} CFU/g; \(P < 0.01\)), indicating that SPRAB is a suitable recovery medium for the enumeration of heat-stressed *E. coli* O157:H7 in ground beef patties. In contrast, no significant difference between counts obtained with SMAC and those obtained with SPRAB was observed for uncooked ground beef patties (control).

To validate the safety of the tested cooking methods (71.1°C with no holding time) with regard to *E. coli* O157:H7 in ground beef patties, patties were inoculated with lower levels (<10^4 CFU/g) of *E. coli* O157:H7. Cassin et al. (4) reported that 87% of contaminated ground beef packages were predicted to contain <10^7 *E. coli* O157:H7 cells, and packages containing 10 to 10^4 *E. coli* O157:H7 cells were predicted to be rare, accounting for the remaining 13% of the contaminated packages. Additionally, <10^3 CFU of *E. coli* O157:H7 per g has been isolated from ground meat (2). Therefore, the 10^3-CFU/g level was deemed appropriate for the evaluation of the safety of ground beef cooked to an internal temperature of 71.1°C with no holding time. After inoculation and cooking, *E. coli* O157:H7 counts on SMAC agar were determined for directly plated samples and for enriched samples. No *E. coli* O157:H7 was found in ground beef patties cooked by the MT-SSG or the DSG method (Table 3). However, two samples (22%) of ground beef cooked by the OT-SSG method tested positive after enrichment, even though no *E. coli* O157:H7 was detected by direct plating. The OT-SSG method resulted in an *E. coli* O157:H7 reduction of 4.7 log_{10} CFU/g when high levels (>10^7 CFU/g) of *E. coli* O157:H7 were used in the first experiment (Fig. 1). However, if beef patties have lower levels of *E. coli* O157:H7 (3 to 4 log_{10} CFU/g) and are cooked by the OT-SSG method, they still pose a potential risk of food poisoning, likely because of the increased heat resistance due to heat shock during the slower OT-SSG cooking process. *E. coli* O157:H7 can adapt to conditions of sublethal heating, enabling subsequent survival of the organism at more lethal temperatures (9, 22, 28). As the cell experiences a slow increase in temperature, the production of heat shock proteins is accelerated in a very short time. Yamamori et al. (32) found that the rate of synthesis of five cellular proteins in *E. coli* increased 5- to 10-fold within 5 min after a temperature increase. Moreover, other factors with regard to ground beef patties, such as air pockets and different fat distributions, may contribute to the different heat resistance levels of *E. coli* O157:H7.

In conclusion, the more rapid cooking method was more effective in destroying *E. coli* O157:H7 in ground beef patties. The reduction rate achieved (Fig. 1) was associated with the cooking time required to reach a target temperature (Table 1). If a cooking method results in very little within-patty temperature variation, the end point target temperature of 71.1°C without a holding time is adequate. However, ground beef patties in a skillet do not cook uniformly in a given time at a given temperature. Moreover, different cooking systems or cooking methods may result in differences in postcooking heat retention for patties. The wide variation in within-patty temperatures and the detection of heat-injured *E. coli* O157:H7 in patties cooked on an electric grill and turned only once indicates that there is some risk of pathogens surviving the cooking process with the OT-SSG method. Consumers should be advised that cooking ground beef patties in a double-sided grill or turning patties frequently (every 30 s) when cooking on a single-sided grill results in a more uniform temperature and more reliable destruction of pathogens. In addition, educational campaigns with regard to the recommended end point temperature and the need to take multiple temperature readings for each ground beef patty are essential. Additional investigation of other cooking methods or systems available to consumers is needed to ensure the safety of cooked ground beef patties.

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