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Thermal Inactivation of H5N1 High Pathogenicity Avian Influenza Virus in Naturally Infected Chicken Meat

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ABSTRACT

Thermal inactivation of the H5N1 high pathogenicity avian influenza (HPAI) virus strain A/chicken/Korea/ES/2003 (Korea/03) was quantitatively measured in thigh and breast meat harvested from infected chickens. The Korea/03 titers were recorded as the mean embryo infectious dose (EID_{50}) and were $10^{8.0}$ EID_{50}/g in uncooked thigh samples and $10^{7.5}$ EID_{50}/g in uncooked breast samples. Survival curves were constructed for Korea/03 in chicken thigh and breast meat at 1°C intervals for temperatures of 57 to 61°C. Although some curves had a slightly biphasic shape, a linear model provided a fair-to-good fit at all temperatures, with R^2 values of 0.85 to 0.93. Stepwise linear regression revealed that meat type did not contribute significantly to the regression model and generated a single linear regression equation for *z*-value calculations and *D*-value predictions for Korea/03 in both meat types. The *z*-value and the upper limit of the 95% confidence interval for the *z*-value were 4.64 and 5.32°C, respectively. From the lowest temperature to the highest, the predicted *D*-values and the upper limits of their 95% prediction intervals (conservative *D*-values) for 57 to 61°C were 241.2 and 321.1 s, 146.8 and 195.4 s, 89.3 and 118.9 s, 54.4 and 72.4 s, and 33.1 and 44.0 s. *D*-values and conservative *D*-values predicted for higher temperatures were 0.28 and 0.50 s for 70°C and 0.041 and 0.073 s for 73.9°C. Calculations with the conservative *D*-values predicted that cooking chicken meat according to current U.S. Department of Agriculture Food Safety and Inspection Service time-temperature guidelines will inactivate Korea/03 in a heavily contaminated meat sample, such as those tested in this study, with a large margin of safety.

High pathogenicity avian influenza (HPAI) viruses cause severe disease with high mortality in chickens and related gallinaceous poultry. In chickens, the initial replication of the HPAI virus occurs in the respiratory or intestinal tract and is followed by systemic spread of the virus through the blood, with subsequent infection of internal organs, brain, skin, and skeletal muscle *(1, 13, 17, 22, 28).* As reviewed by Swayne and Pantin-Jackwood *(24),* most AI virus strains that exhibit high pathogenicity in chickens do not cause severe disease in domestic ducks. However, some of the 2001 through 2006 H5N1 HPAI viruses have caused systemic disease in ducks with varying levels of mortality. The presence of HPAI virus in the meat of infected birds raises the question of whether H5N1 HPAI virus can be transmitted to other poultry, mammals, and humans via contaminated poultry products, either by oral and nasal mucous membrane exposure or by ingestion.

Most cases of H5N1 HPAI virus infection in humans have been linked to direct contact with diseased birds *(4, 8, 16, 20, 38).* Although consumption of undercooked or raw products from infected birds have been implicated as a possible mechanism for H5N1 HPAI virus exposure in a few cases, to date no conclusive epidemiological evidence has supported foodborne transmission of the virus to humans. As summarized by Butler *(5),* whether the gastrointestinal tract can act as a portal of entry for the virus remains a topic of debate. However, field observations and experimental studies clearly indicate that H5N1 HPAI virus infections in birds, mammals, and humans can have an enteric component.

Because of the severe nature of HPAI in poultry and the possibility of transmission to humans, the World Organization for Animal Health (Office International des Epizooties) recommends that poultry products from countries, zones, or compartments where HPAI virus has been found be treated to inactivate the virus prior to export *(14).* The demonstration of heat inactivation of AI virus in poultry products suggests that thermal processing could be an effective treatment method *(19, 21).* A preliminary study performed in our laboratory involved a precise microassay system developed for measuring thermal inactivation of AI virus in meat samples, but quantitative measurements of AI virus inactivation were not made at that time *(19).* In the present study, we obtained quantitative measurements of heat sensitivity for a representative H5N1 HPAI virus strain in chicken meat and evaluated the efficacy of current U.S. Department of Agriculture Food Safety and Inspection Service (USDA FSIS) cooking guidelines *(32)* with respect to H5N1 HPAI virus inactivation.

MATERIALS AND METHODS

Virus inoculum. Working stocks of the H5N1 virus strain A/chicken/Korea/ES/2003 (Korea/03) were the second passage grown in 11-day-old embryonating chicken eggs. Amnioallantoic fluid was harvested 30 to 48 h after allantoic sac inoculation and

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diluted to a mean embryo infectious dose (EID_{50}) of 10^6 per 0.1 ml in the protein-rich buffered medium Bacto brain heart infusion (BHI; Becton Dickinson, Sparks, Md.). All work with the virus and infected material was performed in USDA-certified biosafety level 3 agriculture facilities.

Animal experimental design. Five 4-week-old specific-pathogen-free White Leghorn chickens were each inoculated intranasally with 10^6 EID_{50} of Korea/03 virus in 0.1 ml of BHI. All five chickens were found dead at 2 days postinoculation. Thigh and breast tissue were collected and stored at −70°C. Chickens were housed in negative pressure high-efficiency particulate air ventilated stainless steel isolation cabinets under constant illumination. Food and water were provided as needed.

Thermal inactivation procedure. Samples of thigh and breast meat from one chicken were used for all thermal inactivation experiments. Thermal inactivation was performed as described previously *(19),* except that samples were heated in a GeneAmp 9700 thermocycler (Applied Biosystems, Foster City, Calif.). Raw skinless meat samples (0.05 ± 0.002 g) were placed in thin-walled polypropylene PCR tubes and centrifuged to pack the samples into the bottom of the tubes. Triplicate meat samples were prepared for most time points, but in a few cases four or five meat samples (duplicate runs) were tested. Samples were placed in the thermocycler heating block at 25°C and removed after treatment for the specified length of time at the target temperature. For the zero time point at each temperature, samples were removed immediately after the heating block reached the target temperature. All samples were maintained at 4°C before treatment and were chilled in a 4°C cold block (CoolSafe, Diversified Biotech, Boston, Mass.) immediately upon removal from the thermocycler.

Virus isolation and titration. Virus isolation and titration were performed as described previously *(19).* Heat-treated meat samples were allowed to chill at 4°C, transferred to 1.7-ml polypropylene centrifuge tubes, and ground with 0.5-ml pestles, and 0.5 ml of BHI containing appropriate antibiotics was added to each ground meat sample. The resulting 10% tissue suspension was vortexed and then centrifuged. A 0.1-ml aliquot of supernatant was inoculated into each of three 9- to 11-day-old embryonating chicken eggs for virus isolation and titration *(25).* The 50% endpoints were calculated using the method of Reed and Muench *(36),* and virus titers were recorded as log EID_{50} per gram of meat. The detection limit of the assay was $10^{2.2}$ EID_{50}/g.

Statistics and graphs. Statistical operations were performed with Sigma Stat version 2.03 (1992 through 1997, SPSS, Chicago, Ill.). Graphs were prepared with Sigma Plot (2000, SPSS). The distribution of the virus titer data was approximately lognormal (the mean was approximately equal to the median) and fulfilled the normality requirement for parametric statistical tests. Experimental *D*-values were calculated from linear regression of virus titer versus time at the given temperature (*D*-value = −1/slope). The upper limit of the 95% confidence interval for the slope coefficient was used to calculate a conservative experimental *D*-value for each temperature. The following equation was used to calculate the upper limit of the 95% confidence interval for the slope coefficient:

$$b_1 + t^*(s_\varepsilon)$$

where b_1 is the slope coefficient, t^* is obtained from a *t* test critical values table (two-tailed test, $\alpha = 0.05$), and s_ε is the standard error of the slope coefficient. The *z*-values for thigh and breast meat were calculated from linear regressions of log *D*-values (seconds) versus temperature (*z*-value = −1/slope). The upper limit of the 95% confidence interval for each *z*-value was calculated as described for the *D*-values, except that the following generic equation was used to calculate the upper limit of the 95% confidence interval for the slope coefficient:

$$b_1 + 2(s_\varepsilon)$$

For *D*-values calculated from a regression line equation, the following equation was used to calculate the upper limits of the 95% prediction intervals:

$$y + 2(\text{RMSE})$$

where *y* is the predicted log *D*-value (seconds) and RMSE is the root mean square error or the standard error of the *y* estimate.

RESULTS

Amount of H5N1 HPAI virus present in meat from infected chickens and ducks. For this study, triplicate samples of raw chicken meat were assayed to determine the amount of Korea/03 virus present. Titers for thigh meat ranged from $10^{7.8}$ to $10^{8.5}$ EID_{50}/g, with an average of $10^{8.0}$ EID_{50}/g. The titer for each of the three breast meat samples was $10^{7.5}$ EID_{50}/g. Virus titers in meat from H5N1 HPAI–infected chickens and ducks during other studies are listed in Table 1. Comparison with other chicken meat samples infected with H5N1 HPAI virus isolates and evaluated in our laboratory revealed that the average titers in this study may not be unusually high *(3, 19, 22, 28).* H5N1 HPAI virus also can be found in the skeletal muscle of infected ducks, even before clinical signs of HPAI virus infection appear *(3, 15).*

Survival curves and *D*-values for Korea/03 virus in chicken meat. Figure 1 shows survival curves for Korea/03 virus in chicken thigh and breast meat at temperatures ranging from 57 to 61°C in 1°C intervals. Although some of the curves had a slightly biphasic shape, a linear model provided a fair-to-good fit for all curves, with R^2 values of 0.85 to 0.93. None of the curves had shoulders that would indicate lag times for Korea/03 virus inactivation in chicken meat with the skin removed and no added ingredients. Therefore, a linear model was assumed for the purpose of calculating *D*-values (the time required for a 1-log reduction in infectious titer) by linear regression. *D*-values, the upper limits of the 95% confidence intervals for the *D*-values, and the R^2 values for each survival curve are shown in Table 2. In general, similar *D*-values were observed for Korea/03 virus in both meat types.

Calculation of *z*-values and regression line equations. The *z*-value (the temperature increase needed to reduce the *D*-value by 1 log) describes the temperature dependence of a thermal inactivation reaction. A regression plot of log *D*-value versus temperature yields an equation that can be used to calculate the *z*-value and to predict *D*-values for additional temperatures. Figure 2 shows linear regression plots of log *D*-value versus temperature for Korea/03 virus in chicken thigh and breast meat. Similar regression plots and line equations were obtained for both thigh and breast meat. To determine whether a single line equation could be obtained by combining the *D*-value data,

TABLE 1. *Virus titers in meat from chickens and domestic ducks infected with H5N1 HPAI virus*

Strain	Meat source[a]	Titer (log EID_{50}/g)	Clinical status of infected bird[b]	Reference
A/chicken/Korea/ES/03	Chicken thigh	8.0[c]	Dead	This study
A/chicken/Korea/ES/03	Chicken thigh	6.8[c]	Dead	*19*
A/chicken/Korea/ES/03	Chicken breast	7.5[c]	Dead	This study
A/chicken/Korea/ES/03	Chicken breast	5.6[c]	Dead	*19*
A/chicken/Korea/ES/03	Chicken breast	7.3[d]	Dead	*22*
A/duck/Anyang/AVL-1/01	Chicken breast	5.5[e]	Dead or sick	*28*
A/Indonesia/05/05	Chicken breast	7.9[f]	Dead	*3*
A/duck/Anyang/AVL-1/01	Duck thigh	3.4[f]	Clinically normal	*3*
A/Env/Hong Kong/437-6/99	Duck thigh	2.0[f]	Clinically normal	*3*
A/Vietnam/1203/04	Duck thigh	5.7[f]	Sick	*15*
A/Prachinburi/6231/04	Duck thigh	4.0[f]	Sick	*15*
A/crow/Thailand/(1C)/04	Duck thigh	5.6[f]	Sick	*15*
A/egret/Hong Kong/757.2/02	Duck thigh	6.0[f]	Sick	*15*
A/egret/Hong Kong/757.2/02	Duck thigh	2.8[f]	Clinically normal[g]	*15*

[a] Chickens were 3- to 4-week-old White Leghorn or White Plymouth Rock. Ducks were 5-week-old White Pekin (last row) or 2-week-old white Pekin (all other rows).
[b] Sick and clinically normal birds were euthanized for tissue collection.
[c] Average titer of three meat samples taken from one infected bird.
[d] Titer of a pooled meat sample from nine infected birds.
[e] Highest breast meat titer reported in the study.
[f] Average titer from two infected birds.
[g] Some of the ducks used for the study were euthanized for tissue collection before clinical symptoms appeared.

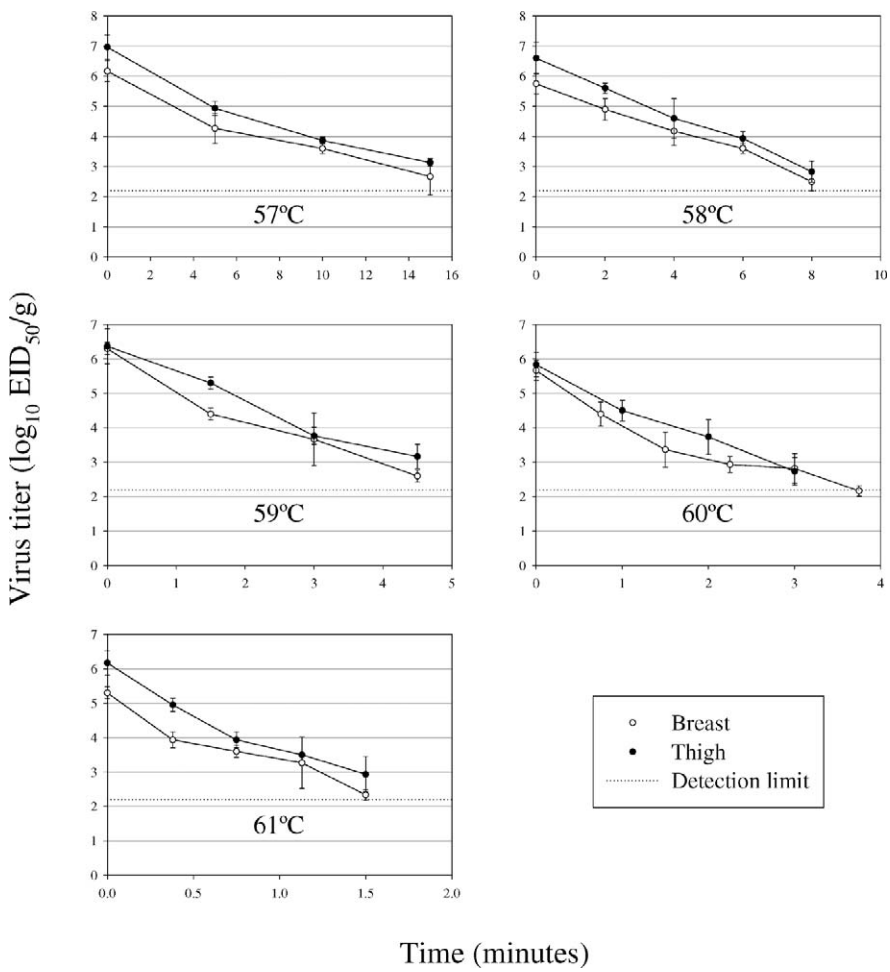

FIGURE 1. *Survival curves for Korea/03 virus in chicken meat. Each data point represents the average titer of at least three meat samples, and the error bars indicate standard deviations. The detection limit of the assay is approximately 2.2 log EID_{50}/g meat.*

TABLE 2. *Experimental* D-*values for Korea/03 virus in chicken meat*

Temp (°C)	Thigh meat			Breast meat		
	D-value (s)[a]	95% UCL (s)[b]	R^2 [c]	*D* value (s)	95% UCL (s)	R^2
57	238.8	296.6	0.93	268.7	364.3	0.88
58	130.4	156.1	0.93	153.8	182.4	0.92
59	80.8	100.4	0.93	76.1	98.6	0.91
60	59.6	76.1	0.91	70.7	89.4	0.85
61	28.6	34.7	0.91	34.1	45.6	0.85

[a] Time calculated from the inactivation curves shown in Figure 1.
[b] 95% upper confidence limit for the *D*-value.
[c] Coefficient of determination.

backward stepwise linear regression was performed. Dummy variables (0 for thigh and 1 for breast) were assigned to the meat types, and terms for both temperature and meat type were included in the initial regression model. Stepwise linear regression analysis revealed that the contribution of meat type to the line equation was not significant ($P = 0.23$) and generated a combined model line equation for *z*-value calculation and *D*-value prediction for Korea/03 virus in chicken meat (Fig. 2). A *z*-value of 4.64°C was calculated from the combined model line equation, and similar *z*-values were calculated from the thigh and breast meat equations (Table 3). The *D*-values predicted by the combined model line equation are shown in Table 4. To account for error in the model's ability to predict *D*-values, conservative *D*-value estimates were obtained by calculating the upper limits of the 95% prediction intervals for the *D*-values.

Reduction of Korea/03 virus titer during thermal processing of chicken meat. A conservative but realistic H5N1 HPAI virus titer for a heavily contaminated chicken meat sample was estimated by adjusting the highest H5N1 HPAI virus titer reported in Table 1 ($10^{8.0}$ EID_{50}/g) for variability in titer measurement. Because the titer data (log scale) have a normal distribution, approximately 97% of titers for a given meat sample are expected to fall within 2.2 standard deviations of the average titer for that sample. For triplicate meat samples evaluated for this study, the average standard deviation from the mean titer was $10^{0.32}$ EID_{50}/g. Therefore, adding 2.2 standard deviations to the highest average titer listed in Table 1 yields a titer of $10^{8.7}$ EID_{50}/g. Based on our current knowledge of H5N1 HPAI virus concentrations in chicken meat (Table 1), this value can be considered a representative for a high-titer meat sample.

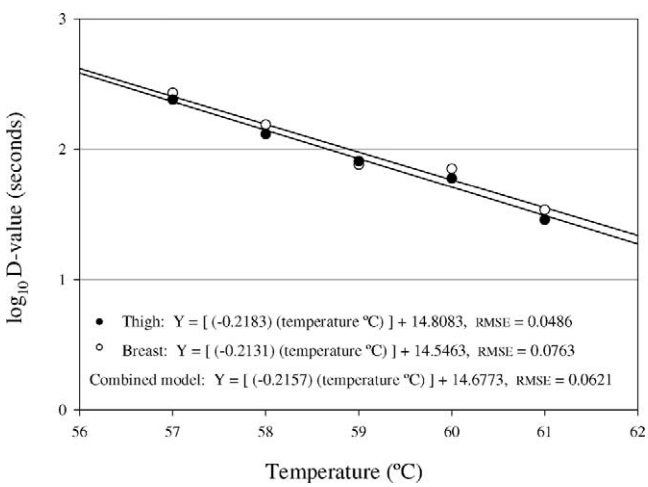

FIGURE 2. *Line equations and linear regression plots of log* D-*value (in seconds) versus temperature (in °C) for Korea/03 virus in chicken thigh and breast meat. The combined model line equation for predicting* D-*values in both meat types was obtained by stepwise linear regression. RMSE, root mean square error.*

TABLE 3. z-*values for Korea/03 virus in chicken meat*

Line equation	*z*-value (°C)	95% UCL (°C)[a]	R^2 [b]
Thigh	4.58	5.33	0.99
Breast	4.69	6.07	0.96
Combined model	4.64	5.32	0.97

[a] 95% upper confidence limit for the *z*-value.
[b] Coefficient of determination.

Calculations were performed to determine whether current USDA FSIS time-temperature guidelines for cooking chicken meat to achieve a 7-log reduction of *Salmonella (32)* are also sufficient for the inactivation of high titers of Korea/03 virus in a 100-g (cooked) serving of meat. Based on the FSIS assumption of a 70% yield by weight after cooking *(30),* 143 g of raw meat would yield 100 g of cooked product. Our representative high-titer meat sample would have approximately $10^{10.9}$ EID_{50} of Korea/03 virus in 143 g of meat. Therefore, an 11-log reduction process would be expected to destroy all of the infectious Korea/03 virus particles present in the uncooked serving of meat.

To ensure that the target lethality for Korea/03 virus was met, predictions for Korea/03 virus inactivation were based on the upper limits of the 95% prediction intervals for the *D*-values. An 11-log reduction in Korea/03 virus titer in chicken meat would be achieved well before the minimum FSIS time-temperature guidelines were met (Table 5), and many additional log reductions of Korea/03 virus would be expected if the internal target temperature of the meat were maintained for the full time specified by the guidelines.

TABLE 4. D-*values for Korea/03 virus in chicken meat calculated from the combined model line equation shown in Figure 2*

Temp (°C)	Predicted *D*-value (s)	95% PI upper limit[a]
57	241.2	321.1
58	146.8	195.4
59	89.3	118.9
60	54.4	72.4
61	33.1	44.0

[a] Upper limit of the 95% prediction interval for the *D*-value.

TABLE 5. *Time predicted for an 11-log reduction of Korea/03 virus titer in chicken meat at a given internal temperature and number of log reductions of Korea/03 virus titer achieved in chicken meat cooked according to minimum current USDA FSIS time-temperature guidelines for a 7-log reduction of* Salmonella

Temp		95% PI upper limit for *D*-value (s)[a]	Time predicted for an 11-log EID_{50} reduction of Korea/03 virus titer	Minimum FSIS time-temp guideline[b]	Predicted no. of log EID_{50} reductions of Korea/03virus/ achieved[c]
°C	°F				
57.8	136	215.8	39.6 min	63.3 min	17.6
58.9	138	125.0	22.9 min	39.7 min	19.1
60.0	140	72.4	13.3 min	25.2 min	20.9
61.1	142	41.9	7.7 min	16.1 min	23.1
70.0	158	0.50	5.5 s	21.9 s	43.8
73.9	165	0.073	0.80 s	<10 s[d]	13.7/s

[a] Upper limit of the 95% prediction interval for the *D*-value, calculated from the combined model line equation + 2RMSE (Fig. 2). All of the predictions in Table 5 are conservative estimates based on this number. RMSE, root mean square error.
[b] From the time-temperature table for chicken meat with 1% fat.
[c] Assuming that the required internal temperature is maintained for the length of time specified in the FSIS time-temperature table.
[d] Required lethality is achieved instantly at this internal temperature.

DISCUSSION

To date, no conclusive epidemiological evidence has supported foodborne transmission of the H5N1 HPAI virus to humans. However, field observations and experimental studies clearly indicate that H5N1 HPAI virus infections in animals and humans can have an enteric component. Captive tigers and leopards were infected during a recent H5N1 HPAI outbreak in Thailand after consuming raw chicken carcasses presumed to be infected with the virus *(10, 26).* During an H5N1 HPAI outbreak in wildlife on Ruegen Island, Germany, in early 2006, domestic cats and a stone marten (a carnivore belonging to the weasel family) were infected, presumably after eating infected birds *(37).* Experimentally, oral transmission has been demonstrated by feeding H5N1 HPAI virus–infected chicks to domestic cats *(12, 18).* Chickens can be infected with H5N1 virus by eating contaminated meat, provided that the virus titer in the meat is sufficiently high *(22).* In humans, epidemiological studies and case reports provide evidence for enteric involvement in at least some cases of H5N1 HPAI virus infection. Gastrointestinal symptoms such as diarrhea were commonly observed in H5N1 HPAI virus–infected patients during recent outbreaks in Southeast Asia *(6, 27),* and a few patients have demonstrated gastrointestinal symptoms in the absence of respiratory symptoms *(2, 7).* Taken together, the current evidence for enteric involvement of the H5N1 HPAI virus suggests that the possibility of foodborne transmission merits further scientific investigation.

Viruses normally implicated in outbreaks of foodborne illness, such as hepatitis A virus and the Noroviruses, are relatively heat resistant and highly infectious *(11).* In contrast, the AI virus strains tested in poultry products to date have been relatively heat sensitive, and differences in heat sensitivity among AI virus strains are not expected to be large because of the similar physical and chemical properties of these viruses *(23).* Because currently circulating H5N1 HPAI virus strains are not easily transmitted to humans, the infectious dose for humans is expected to be relatively high regardless of the route of exposure. However, the infectivity of H5N1 HPAI virus for humans could change if strains that are better adapted to a human host emerge. A complete risk assessment for foodborne transmission of H5N1 HPAI virus to humans would require consideration of additional factors, such as the probability of infected birds entering the food chain undetected and the risk of recontamination of poultry products with H5N1 HPAI virus during processing. The current study addresses one aspect of risk by providing quantitative thermal inactivation data for a representative H5N1 HPAI virus strain in chicken meat. These data can be used to evaluate the efficacy of thermal processing schedules for H5N1 HPAI virus inactivation in chicken meat as shown in Table 5 for current USDA FSIS cooking guidelines.

The USDA FSIS performance standard for processing ready-to-eat poultry products specifies acceptable probabilities for the survival of *Salmonella* such that the finished product poses no health risks to consumers, even with a worst-case raw product *(31, 33).* Processing schedules that meet this standard are, in most cases, expected to result in the destruction of other foodborne pathogens in poultry meat. The FSIS provides time-temperature guidelines *(32)* that meet the performance standard for a 7-log reduction of *Salmonella* in chicken and turkey *(9).* Because the lag time for thermal inactivation of *Salmonella* in poultry meat increases with increasing fat content *(9),* the FSIS recommends longer cooking times for meat with higher fat percentages. Separate time-temperature guideline tables are provided (in 1% increments) for poultry meat with 1 to 12% fat.

According to van Asselt and Zwietering *(35),* lower water activity and higher fat content in food products are generally expected to provide some protection to bacterial pathogens during thermal processing. Likewise, the specific qualities of the heating menstruum have been shown to affect the thermal inactivation of foodborne viruses *(11).* The exact fat content of the meat samples used in the current study is unknown but probably was relatively low because the skin was removed and no additional ingredients were added. The USDA *Nutrient Database for Standard Reference* lists average fat contents of 3.91 and 1.24% for raw,

skinless chicken thigh and breast meat, respectively, from broilers and fryers *(29).* Lag times and significant differences in Korea/03 virus inactivation in thigh versus breast meat were not detected in the current study (Figs. 1 and 2). However, the possibility that higher fat content could affect the processing time required for H5N1 HPAI virus inactivation should be considered.

A simple regression line equation that incorporates the *D*-value data from both meat types was used to predict thermal inactivation of Korea/03 virus in raw, skinless chicken meat at different temperatures (Table 5). Both 70 and 73.9°C are temperatures of interest to industry and consumers because safe cooking guidelines aimed at consumers recommend cooking chicken meat to these internal temperatures *(34, 38).* The calculations shown in Table 5 suggest that an 11-log reduction in Korea/03 virus titer should take place in less than 1 s at 73.9°C but could take 5.5 s at 70°C. The *D*-values for 70 and 73.9°C were predicted by extrapolation beyond the range of temperatures included in this study, and the true accuracy of the regression model in this range is unknown. However, these 70°C calculations do not contradict results from previous work *(19).* In that study, 1 s at 70°C was not always sufficient for reducing Korea/03 virus titers to below the detection limit of the assay ($10^{2.2}$ EID_{50}/g), but Korea/03 virus was not detected in any sample treated at 70°C for 5 s.

The full 95% confidence interval for the 70°C *D*-value is 0.28 to 0.50 s. Therefore, only a 2.0- to 3.5-log reduction in Korea/03 virus titer would be expected after 1 s at 70°C. The total log reduction of Korea/03 virus achieved during the entire cooking process would actually be greater because a significant amount of virus is inactivated during heating to 70°C *(19),* and additional loss would be expected during the cooling stage of the cooking process. The *D*-values presented in this study, combined with data on heat transfer through a meat sample during thermal processing, would allow establishments to estimate the total lethality of a processing schedule for H5N1 HPAI virus. When the total lethality of a process for H5N1 HPAI virus is unknown, cooking to an internal temperature of 73.9°C or ensuring that the internal temperature of the meat remains at 70°C for 5 s are safer options for H5N1 HPAI virus inactivation.

The 11-log reduction example is not meant to be a proposed lethality performance standard for H5N1 HPAI virus analogous to the 7-log performance standard for *Salmonella* reduction because the estimate of $10^{8.7}$ EID_{50}/g for a representative sample with a high H5N1 HPAI virus titer is uncertain. The highest H5N1 HPAI virus titers in meat were obtained from dead birds (Table 1), which would not enter the human food chain because they would be rejected at the processing plant. The concentration of H5N1 HPAI virus that can be present in the meat of infected but asymptomatic or mildly ill chickens is unknown but is expected to be lower than H5N1 HPAI virus concentrations in meat from infected dead chickens. In addition, the H5N1 HPAI virus titer data are based on a small data set (Table 1), and the amount of virus lost during extraction from the meat of infected birds is unknown. However, the calculations in the last column of Table 5 show that cooking chicken meat according to the FSIS time-temperature guidelines provides a large margin of safety for Korea/03 virus inactivation due to the large number of log reductions expected when the target internal temperature of the meat is maintained for the specified time, and additional log reductions are expected during the heating and cooling stages of the cooking process.

Because HPAI is a zoonotic disease that is present in the tissues of infected birds, strategies to keep HPAI virus out of the food supply should emphasize control measures for reducing the spread of all AI viruses in poultry. The risk of infected birds entering the human food chain undetected can be minimized by implementing comprehensive AI control programs, which include strict biosecurity measures, surveillance and detection programs, and poultry elimination when appropriate.

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