***Salmonella enteritidis* outbreak in a restaurant chain: the continuing challenges of prevention**

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SUMMARY

In 1990, a *Salmonella enteritidis* (SE) outbreak occurred in a restaurant chain in Pennsylvania. To determine its cause(s), we conducted a case-control study and a cohort study at one restaurant, and a survey of restaurants. Egg dishes were associated with illness ($P = 0.03$). Guests from one hotel eating at the restaurant had a diarrhoeal attack rate of 14%, 4.7-fold higher than among those not eating there ($P = 0.04$). There were no differences in egg handling between affected and unaffected restaurants. Eggs supplied to affected restaurants were medium grade AA eggs from a single farm, and were reportedly refrigerated during distribution. Human and hen SE isolates were phage type 8 and had similar plasmid profiles and antibiograms. We estimate the prevalence of infected eggs during the outbreak to be as high as 1 in 12. Typical restaurant egg-handling practices and refrigeration during distribution appear to be insufficient by themselves to prevent similar outbreaks.

INTRODUCTION

In the United States, the number of reported human isolates of *Salmonella enteritidis* (SE) has been increasing steadily since the early 1980s, first in the Northeastern and mid-Atlantic states and more recently in other parts of the country [1–3]. In 1990, SE became the most frequently reported salmonella serotype in the Centers for Disease Control Salmonella Surveillance Annual Summary [4]. Concomitantly, the number of outbreaks of SE gastroenteritis has risen throughout the Northeast and spread to states outside this region [3]. This increase is attributable to contaminated grade A shell eggs. Eggs are graded AA, A, B in decreasing order of quality by US standards. From 1985–9, shell eggs accounted for 82% of the outbreaks in which a food vehicle for transmission was identified [3]. In February 1990, the US Department of Agriculture (USDA) initiated a control program based on tracing eggs from disease outbreaks to source egg farms, testing the internal organs of chickens on those farms, and diverting the eggs to pasteurization if cultures yield SE [5]. In April 1990, the US Food and

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Drug Administration (FDA) recategorized eggs as a potentially hazardous food requiring proper refrigeration and cooking [6]. Some states are considering legislation to mandate refrigeration of eggs and use of pasteurized egg products in institutions to decrease outbreaks of SE infections.

We recently investigated a SE gastroenteritis outbreak associated with a restaurant chain in Pennsylvania, which illustrates the issues that continue to challenge prevention of such outbreaks.

MATERIALS AND METHODS

Background

In early July 1990, routine surveillance and case-investigation in Pennsylvania identified several persons with laboratory-confirmed SE gastroenteritis who reported having eaten at different chain A restaurants 1–2 days before symptom onset. The Pennsylvania Department of Health (DOH) analysed open-ended food histories collected from all reported case-patients in a case-control fashion, using healthy meal companions as controls, and found egg consumption to be significantly associated with illness.

Case-control investigation

To identify the food vehicle of transmission more precisely, we carried out a case-control investigation at the chain A restaurant with the greatest number of cases reported. We defined a case as documented infection with *Salmonella* serotype *enteritidis* in a person who had eaten at this restaurant within 7 days before onset of gastrointestinal symptoms. Case-patients were administered a telephone questionnaire on symptoms, meals eaten at the restaurant, and other exposures. We then conducted similar interviews of meal companions; those with gastrointestinal symptoms in the week following their meal were excluded from the control group.

Cohort investigation

To assess attack rates, we reviewed records of all guests staying at a hotel adjacent to the above restaurant from 30 June through 9 July. We administered to all guests reachable by telephone a questionnaire which asked about symptoms, history of eating at the implicated restaurant, food history if appropriate, and other exposures. A case was defined as gastrointestinal illness with diarrhoea (three or more loose stools in a 24-h period) in a guest beginning during or within a week after staying at this hotel.

Survey of restaurants

To determine the role of food-handling deficiencies contributing to the outbreak, we then conducted a restaurant case-control study. A restaurant was defined as affected if two or more unrelated culture-confirmed cases of SE detected by routine surveillance reported they ate there in the week before onset, and had symptom onset during the period 15 June to 22 July, 1990. Nine restaurants of this chain were affected. For one comparison group, we selected the 11 other restaurants of the same chain in Pennsylvania which received eggs from the same farm as the affected restaurants but which were not affected. As a second

comparison group, we selected 16 other restaurants not belonging to this chain which received their eggs from other sources and which also were not affected. These 16 non-chain A restaurants were similar to the affected restaurants: they were also family-style restaurants in the same communities, serving a similar number of patrons and similar menus, including all major egg dishes. At each site, we interviewed a manager, knowledgeable restaurant representative, or cook using a questionnaire on egg delivery, storage, handling, and cooking techniques practiced before the outbreak. Efforts were made to elicit details concerning egg handling before the outbreak by conducting face-to-face anonymous interviews. We also reviewed past county and corporate restaurant inspection scores as an objective surrogate of the restaurants' past overall food-handling performance. Affected restaurants were inspected by local health department sanitarians.

Trace-back investigation

To trace eggs delivered to affected restaurants, receipts and invoices from affected restaurants and food distributors were reviewed. We visited food distributors to verify that storage and transport facilities were refrigerated. The USDA investigated the source farm and sampled the hens. The USDA and the Pennsylvania Department of Agriculture (PDA) also visited the pullet farm that raised the implicated flock and collected environmental samples of chicken litter, cages, feed belts, and ventilation fans for culture.

Laboratory investigations

Human stool specimens were processed by the Pennsylvania DOH Laboratory in Lionville, Pennsylvania. Chicken necropsy specimens were processed at the National Veterinary Services Laboratories in Ames, Iowa. Hens' organs were processed in a stomacher, pooled, enriched in tetrathionate broth, incubated at 37 °C for 24 h, then plated onto MacConkey agar and brilliant green agar with Novobiocin. Environmental specimens from the pullet farm were cultured at the Lionville DOH laboratory and the PDA Sunnydale laboratory following similar enrichment and culture procedures. Salmonella isolates were identified and serotyped following standard laboratory procedures [7]. Ten representative SE isolates from humans and all SE isolates from hens were tested at CDC for antibiotic susceptibilities and phage typed using the method of Ward and colleagues [8]; plasmid profiles were determined using a modified Birnboim technique [9].

Statistical analysis

Chi-square analysis with Yates' correction or Fisher's exact test was used for binomial variables. The Student's *t* test or Wilcoxon rank sum test was employed for continuous variables. Ninety-five percent confidence limits were calculated for odds ratios (OR) [10] and for relative risks (RR) [11]. All *p*-values are 2-tailed unless otherwise specified.

RESULTS

Descriptive epidemiology

Pennsylvania DOH notifiable disease surveillance identified 94 persons with illness and exposure to nine chain A restaurants; of these, the 79 culture-confirmed

Fig. 1. (a) Culture-confirmed cases of *Salmonella enteritidis* infection by date of onset, Pennsylvania, June-July 1990. (b) Culture-confirmed cases of *Salmonella enteritidis* infection by date of onset, Pennsylvania, June-July 1990.

Fig. 2. (a) Culture-confirmed cases of *Salmonella enteritidis* infection by date of implicated meal, Pennsylvania, June–July 1990. (b) Culture-confirmed cases of *Salmonella enteritidis* infection by date of implicated meal, Pennsylvania, June–July 1990.

Table 1. *Selected characteristics of all patients and of persons from the case-control study, Pennsylvania, June-July 1990*

Characteristic	All patients (<i>n</i> = 79)*	Study patients (<i>n</i> = 26)	Study controls (<i>n</i> = 25)
Male sex, no. (%)	20/67 (30)	12 (46)	13 (54)
Mean age in yrs. (Range)	26 (1-71)	31 (3-72)	31 (3-55)
Symptoms, no. (%)			
Nausea	27/56 (48)	23 (89)	
Vomiting	21/58 (36)	13 (50)	
Diarrhoea	60/60 (100)	26 (100)	
Fever	40/54 (74)	22 (85)	
Cramps	48/58 (83)	24 (92)	
Mean incubation period in days (Range)	2.4 (0-7)	1.3 (1-4)	
Consulted physician, no. (%)	69/70 (99)	25 (96)	
Hospitalized, no. (%)	11/59 (19)	5 (19)	
Mean duration in days (Range)	NA†	4.8 (3-6)	

* Variation in denominator frequencies reflects the omission of persons who did not respond to that question.

† NA, Not available.

SE infections are included in this analysis. The first patient had onset on 15 June, cases peaked on 5 July, and the last patient had onset on 22 July (Fig. 1*a*). The epidemic curves for illness onset dates varied between restaurants, lasting several weeks at most (Fig. 1*a, b*). The epidemic curves for dates of implicated meals also varied between restaurants and were dispersed (Fig. 2*a, b*). The mean age of patients was 26 years (range 1-71 years) (Table 1). The mean incubation period was 2.4 days (range 0-7 days). Diarrhoea was reported by 100%, fever by 74%, and cramps by 83% of the involved patients. Nineteen percent were hospitalized and no deaths were reported. Two persons were reported to have undergone appendectomy because of their clinical symptoms, and one woman reportedly delivered an infant with postpartum *Salmonella* infection.

Case-control study results

Twenty-six (90%) of 29 persons with reported culture-confirmed cases associated with this restaurant (Restaurant 2 in Figs. 1 and 2) were contacted. All had onset of symptoms between 1 and 18 July, with a peak between 5 and 7 July. Except for nausea, these 26 did not differ significantly from the larger group of case-patients from all nine restaurants in age, sex, symptoms, and severity of illness (Table 1). Dishes with eggs as the principal ingredient were the only foods significantly associated with illness (OR = 4.5, 95% CI = 1.1-18.9, *P* = 0.03) and the only foods consumed by more than half of the case-patients (Table 2). No specific egg dish was significant by itself. Pancakes, which had some eggs in its batter and which were reportedly cooked thoroughly until dry, were not significantly associated with illness. Illness was not associated with other potential exposures outside the restaurant. Twenty-five case-patients ate at this restaurant only once in the week before onset. Among these, the mean time from the meal to disease onset was 1.3 days (range 1-4 days).

Table 2. Associations between illness and foods eaten at one chain A restaurant, Pennsylvania, June-July 1990

Food	No. (%) case-patients exposed (n = 26)	No. (%) controls exposed (n = 25)	P
Milk	5 (19)	1 (4)	NS†
Cheese	13 (50)	8 (32)	NS
Salad	7 (26)	2 (8)	NS
Soup	1 (4)	0 (0)	NS
Chicken	4 (16)	3 (12)	NS
Turkey	2 (8)	1 (4)	NS
Ham	6 (23)	3 (12)	NS
Bacon	7 (27)	3 (12)	NS
Sausage	2 (8)	2 (8)	NS
Beef	2 (8)	6 (24)	NS
Pancake	9 (35)	5 (20)	NS
Egg dishes	18 (75)	10 (40)	0.03
Scrambled	7 (29)	3 (12)	NS
Easy over	2 (8)	2 (8)	NS
Omelet	6 (25)	3 (12)	NS
Benedict	0 (0)	0 (0)	NS
French toast	3 (12)	2 (8)	NS

* Denominator may vary reflecting the omission of persons who did not respond to that question.

† NS. Not significant.

Cohort investigation results

We interviewed 148 hotel guests who were at the hotel between 30 June and 9 July; this represented 28% of the registered guests during that time. Four of the 148 had non-diarrhoeal gastrointestinal symptoms and were excluded from analysis. Twelve (8%) of the remaining 144 guests reported symptoms meeting the case definition. All had diarrhoea, 83% had cramps, 67% had nausea or vomiting, and 58% had fever. Diarrhoeal illness was significantly associated with eating at the adjacent chain A restaurant. Ten (14%) of 74 who ate at this restaurant subsequently had diarrhoea, compared with 2 (3%) of 70 who did not eat there (RR = 4.7, 95%; CI = 1.1-20.8, $P = 0.04$). Diarrhoeal illness began a median of 3 days (range: 1-7 days) after eating. Three of those with diarrhoea consulted a doctor but none had cultures performed for salmonella or were hospitalized. None was a known case-patient. No significant association was found with any one food item.

Restaurant survey results

Within chain A restaurants, no significant differences in reported egg storage and handling procedures were found between the nine affected and 11 unaffected restaurants.

Compared with the 16 non-chain A restaurants, the nine affected chain A restaurants served similar numbers of patrons, but significantly more omelets per day (Table 3). However, no information was available to determine 'doneness' of the omelets served. Overall, no significant differences in reported egg storage and

Table 3. *Selected characteristics of affected chain A vs. non-chain A restaurants, Pennsylvania, June-July 1990*

Characteristic	Affected chain A (<i>n</i> = 9)	Non-chain-A (<i>n</i> = 16)	<i>P</i>
	Mean (S.D.*)	Mean (S.D.)	
Customers/week	4744 (1335)	4160 (1965)	NS†
Cases of eggs/week	20 (7)	15 (14)	NS
French toast orders/day	57 (40)	47 (52)	NS
Scrambled eggs orders/day	172 (98)	131 (110)	NS
Omelet orders/day	138 (84)	44 (28)	0.007
Eggs Benedict orders/day	17 (13)	41 (31)	NS
Inspection score (corporate)‡	88 (6)	87 (7)	NS
Selected egg-handling practices			
Minutes shell eggs left out on counter before use	32 (18)	38 (45)	NS
Proportion using same cooking tool for eggs and other foods	38 %	35 %	NS

* SD, Standard deviation.

† NS, Not significant.

‡ Inspection scores are on a scale of 0-100, with 100 being perfect.

handling procedures were identified. Pasteurized egg products were used for some egg dishes by three unaffected chain A and four non-chain A restaurants. Thus none (0 %) of 9 affected restaurants used pasteurized egg products compared with 7 (26 %) of 27 unaffected restaurants surveyed, although this possible protective effect did not reach statistical significance (OR = 0.26, 95 %; CI = 0.01-2.67, *P* = 0.40).

Only two restaurants, one affected and one unaffected, had numerical scores from local health department inspection during or within the year before the outbreak. On the other hand, prior corporate inspection scores were available for 25 (69 %) of the 36 restaurants. Corporate inspection criteria were not available for comparison; however, they reportedly followed local and state health codes, with 'passing' being a total score of 80 or greater on a scale of 0 to 100. There was no significant difference in the mean corporate inspection scores between affected and unaffected restaurants inside or outside the affected chain.

Affected chain A restaurants used grade AA medium shell eggs in all egg-containing recipes. Raw eggs were not served in any dishes or sauces. No egg delivery route or truck was common to all affected restaurants; the majority had eggs delivered by different trucks on different schedules. Three food workers from two different chain A restaurants were ill with SE infection during the outbreak and were excluded from work until well after their illness was over. One of the three was a case-patient in our case-control study; she was a waitress who ate French toast at work every morning before becoming ill.

Affected restaurants all had fully functional refrigerated storage facilities, and none reported any refrigeration breakdown in the weeks before the outbreak. Local public health sanitation inspectors identified deficiencies at some affected restaurants after the outbreak, including having refrigeration temperatures between 15 and 18 °C, leaving flats of eggs on the counter during usage, and cooking eggs at less than the times recommended by the FDA. Our restaurant

visits also revealed deficiencies at some restaurants, including leaving flats of eggs on the counter at room temperature during usage. However, there were no significant differences in observed deficiencies between affected and unaffected restaurants. Facilities for refrigeration, holding egg batter, and cooking were similar at all restaurants.

Trace-back results

The eggs used by the affected restaurants were traced to a single source farm. Through two food distributors, 63 % of the medium AA eggs from the source farm reached the public through chain A restaurants in this area.

The eggs were reported to be refrigerated after packaging at the farm and during distribution. The food distributors had functioning cold rooms where eggs were stored at air temperatures at or below 7 °C, and functioning refrigeration units on their delivery trucks. No refrigeration malfunctions were reported before onset of the outbreak. The estimated times elapsed in the stages of egg distribution were 1–3 days at the farm after laying, 1–5 days at the first food distributor, 1–2 weeks at the second food distributor, and as long as 7 days at the restaurants.

The egg-producing farm had six henhouses, connected by a common egg conveyor belt to an egg-processing building. Five henhouses contained 80 000 egg-laying hens each; one had recently been depopulated because of senescence. Each henhouse had birds of a different age. The youngest birds, which produced approximately 90 % of the medium sized eggs, had arrived from the pullet farm on 1 May, 1990, at age 20 weeks, and were in the house farthest from the egg-processing building. No lapse in ventilation or power outage had been reported since those hens were housed, and their health and productivity were reported as normal.

The pullet farm was a single building housing approximately 80 000 hens and was reportedly disinfected between each flock.

Laboratory investigation results

At the egg-laying farm, approximately 300 hens (60 from each house) were necropsied. *Salmonella enteritidis* was isolated from internal organs of 16 birds. Eight came from the house with the youngest birds; the other eight included birds from all four other houses. Environmental samples from the pullet farm did not yield salmonella group D.

Review of routine surveillance by the poultry industry and the PDA of the multiplier flock from which the birds in the youngest flock reportedly came revealed one isolate of SE from the environment collected on 16 February, 1990, but no isolates from necropsied hens. It should be noted that multiplier farms are separate from pullet farms and egg producing farms.

All SE isolates from the 10 ill humans and the 16 hens from the source farm had a similar antibiotic susceptibility profile and was susceptible to most antimicrobial agents tested. All were phage type 8, and all except one had a single 36-megadalton plasmid; one human isolate had an additional 28-megadalton plasmid.

Estimated prevalence of infected eggs

We estimated the prevalence of infected eggs causing recognized disease at the peak of the outbreak. At restaurant 2, 5400–6500 eggs were reportedly served

during the peak week of 1 to 8 July. Approximately 15% of the eggs were used in pancakes and French toast, and 85% were individually prepared in orders of 1-3 eggs; thus 4590-5525 ($5400-6500 \times 0.85$) eggs were served in individually prepared orders. In the case-control study, of case-patients who ate eggs, 83% ate one of these individual egg orders; this suggests that of the 26 culture-confirmed cases with onset reported during that week, approximately 22 (26×0.83) resulted from the patient eating a single infected egg. Dividing 22 infected eggs by 4590-5525 eggs served results in a minimum prevalence of 1 in 208 to 1 in 250 eggs infected during this peak week.

The attributable attack rate (AR) for diarrhoea was 11% (14% AR among hotel guests eating at the restaurant minus 3% AR among non-restaurant guests). Applying this rate to the reported 4000 patrons served there during the peak week, we estimate that 440 cases of diarrhoea resulted, most of which would not have been confirmed. Using the same suppositions on individual egg orders, 365 (440×0.83) estimated cases resulted from at least 365 infected eggs, yielding a peak prevalence estimate for egg contamination of 365/4590 to 365/5525, or 1 in 12 to 1 in 15 eggs. These estimates do not take into account contaminated eggs that did not cause disease because they were thoroughly cooked or because they contained few organisms, cases resulting from cross-contamination, or other possibilities. They suggest a very high prevalence of infected eggs occurred briefly in this outbreak.

DISCUSSION

Routine surveillance and case-investigation identified this large and dispersed outbreak of SE gastroenteritis affecting multiple restaurants of a single chain. The dates of exposure to affected restaurants suggest that SE was introduced repeatedly over several weeks. The epidemiologically implicated vehicle of transmission was grade AA medium eggs from a single farm. On the farm, the highest prevalence of SE infection was documented in the flock producing the bulk of medium sized eggs. The SE isolates from infected humans and hens were matched by similar antibiograms, phage type, and plasmid profile.

Recent SE outbreaks have often been caused by dishes made with raw or undercooked eggs, especially grade A shell eggs [1-3, 12-16]. Here, no dishes containing raw eggs were served, the eggs were grade AA, US standards' highest quality, and there was no evidence of systematic undercooking beyond that typical of American cuisine which includes runny yolk on fried eggs and soft-cooked omelets. Holding raw pooled eggs or cooked eggs at improper temperature for a prolonged time has contributed to past outbreaks [2, 12, 17]. However, egg dishes at this chain were generally prepared individually, not by pooling more than 2-3 eggs at a time, and then served almost immediately. We found no evidence of systematic time and temperature abuse in the restaurants; improper handling does not readily explain this outbreak. The restaurant manager interviews indicate that the reported handling of eggs in the affected restaurants was not different from what has always been acceptable in the community. Reported deficiencies at some affected restaurants may have contributed to some individual cases but are not likely to explain the entire outbreak.

It remains possible that egg-handling deficiencies escaped detection. Although we anticipated and attempted to reduce potential information bias on surveys, managers of affected restaurants could have tried to give 'correct' answers. Nonetheless, on-site visits and comparison of restaurant inspection scores suggest that survey responses were not far from the truth. It is also possible that there was an unreported breakdown in refrigeration during distribution, which allowed salmonella to grow to an infectious dose. However, no single lapse or mechanical breakdown in refrigeration could easily explain this outbreak since affected restaurants received their eggs from several trucks on different schedules.

It is clear from laboratory and epidemiologic studies that eggs can be laid already infected with SE despite an intact shell [18–25]. The inoculum size in most of these infected eggs was small [18–20], but heavy SE contamination was demonstrated in some [18]. In this outbreak, many infected eggs probably contained a heavy salmonella dose by the time of consumption, because most ill persons ate only 1–3 eggs in individually prepared orders.

Longitudinal studies of flocks previously associated with SE outbreaks also indicate that the incidence of SE-positive eggs laid by naturally infected hens is typically only 1% [18–20]. The high estimated peak rate of egg contamination in this outbreak suggests that something different happened. The infected eggs were most likely laid by the youngest birds, which also had the highest rate of infection. Although these birds could have acquired their infection vertically, they probably became infected after arriving at the producer farm, since samples from the pullet farm and surveillance at the hatchery did not suggest any likely exposure before then. The infected eggs were laid between the end of May and early June, approximately 1 month after the young birds were introduced into this house. In laboratory flocks exposed to SE, the contamination rates in eggs increase sharply and briefly after SE is introduced. In the first week after initial infection of hens, SE was detected in the contents of up to 73% of the eggs, dropping abruptly to less than 1% by the fourth week post-inoculation [21]. We suggest that a sudden mass contamination, similar to the laboratory flocks exposed to SE, may have occurred in the implicated henhouse, perhaps following exposure to SE already present in the henhouse environment.

If undetected infection of a young chicken flock led to large numbers of infected eggs, how can similar outbreaks among humans be prevented? At the restaurant level, stricter regulation and closer supervision in food handling may decrease the risk, but may require changing the 'community standard' rather than correcting a particular foodhandling error. Salmonella has been recovered from yolks of eggs inoculated with as low as 100 organisms and then boiled for as long as 8 min [26]. Therefore, FDA/USDA cooking guidelines call for cooking eggs until the yolk and white are firm. The public's receptiveness to the hard-cooked egg remains unclear; many Americans prefer to consume their eggs somewhat less firm than is necessary to kill salmonella. Although refrigeration during distribution did not altogether prevent this outbreak, it may have prevented the outbreak from being larger and remains a prudent control measure. Routine refrigeration of eggs is now mandated by state law in Pennsylvania (General Assembly of Pennsylvania Act 32, 8/91).

This outbreak illustrates that not enough is known at the farm level to ensure a ready supply of uninfected eggs. Documenting SE-free flocks at the multiplier

and pullet farms may not be sufficient if the young hens then become infected after reaching the egg farm premises. Methods for cleaning and disinfecting the hen houses and eliminating exposures to SE, and other flock-based control measures need to be evaluated. Further research on the natural history of SE infection in egg production flocks is critical to understanding and, if possible, controlling the problem at the farm level. Currently, if an egg producing farm is found to have SE infected hens, the farmer must choose between diverting the eggs to a pasteurization plant or destroying his hens; either option is likely to result in financial loss.

In the interim, pasteurized egg products provide the most direct preventive approach. These products offer restaurants and institutions a way to continue serving soft-cooked egg dishes with substantially less risk of salmonellosis. No outbreak of salmonellosis in the US has yet been traced to pasteurized egg products. As a result of this outbreak, chain A restaurants have switched to using pasteurized egg products for most egg dishes.

Salmonella enteritidis gastroenteritis due to consumption of infected eggs is a major public health problem in the United States. This investigation underscores the challenges in preventing SE infection outbreaks. Eggs were of the highest quality, grade AA. They were laid by hens that came from a multiplier breeder that passed the current surveillance standard for SE. Refrigeration was reportedly used throughout distribution. Eggs did not appear to be systematically mishandled by affected restaurants, compared with other restaurants in the community. Yet, as many as 1 in 12 eggs may have been heavily infected at the peak of the outbreak. Wider efforts to warn the public of the hazards associated with eggs and to educate them in changing cooking and consumption habits may decrease future outbreaks, but it is imprudent to rely on educational efforts as the only preventive strategy. While efforts to achieve control at the farm level continue to be pursued, widespread use of pasteurized egg products offer a safer alternative for institutions, commercial food establishments, and the public at large.

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