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Inspection and Investigation: Tools for Detecting Sources of Food Contamination and Preventing Illness Outbreaks

George J. Jackson
Former Dean of the Staff College,
Center for Food Safety and Applied Nutrition,
U.S. Food and Drug Administration

Abstract Regulatory agencies such as the U.S. Food and Drug Administration use trained inspectors to monitor the hygienic status of the food supply and of the food production and distribution environments. Inspectors may take samples for laboratory analysis. Investigations are undertaken when standard inspection and analysis do not solve such problems as food contamination or illness outbreaks from unknown sources. These investigations are conducted by teams that may include inspectors, laboratory analysts, and epidemiologists.

Described in detail are two investigations: a food-associated illness outbreak caused by the bacterium *Yersinia enterocolitica* from an unexpected source and a series of illness outbreaks caused by the parasitic protozoa *Cyclospora cayetanensis* that was difficult to detect in food. Investigators must consider that food can become contaminated at all its steps from farmland or fishing waters to the consumer's fork.

There are at least two generalities to remember when discussing microbial foodborne illness. Experience has taught us that there is no strict distinction between so-called foodborne pathogens that infect by way of the alimentary canal and so-called waterborne pathogens that infect the consumer by that route. Although a pathogen's prevalence may differ in water and in food, it may also differ in different types of food. Generally, a waterborne pathogen will ultimately finds its way into food.

A foodborne pathogen is usually easier to detect in an ill patient than in the contaminated food that caused the illness. In food the pathogen tends to be few in number, and in a dormant or even injured state, whereas there are likely to be

many pathogens in an acutely ill patient. The invaders exist in an active growth phase in the ill patient. In food there are competitive microorganisms and varied test inhibitors that make detection difficult, whereas in patients single infections are more common and inhibitors are more constant.

Regulatory agencies, such as the U.S. Food and Drug Administration (FDA), routinely conduct inspections of the food supply and of the food production and distribution environments. These inspections are carried out by trained sanitarians who make observations, take measurements (e.g., pH, temperature, chlorine concentration), and may gather samples for subsequent laboratory analysis. They determine the general safety of the food supply and the general hygiene of the surroundings in which food is handled. For many years they operated on the basis of common sense and historical knowledge about the food supply. More and more, though, the systematic Hazard Analysis/Critical Control Points approach is being implemented in decisions about what, where, and how much to inspect.

Investigations are undertaken when a problem arises, such as a foodborne illness outbreak or a food contamination from an unknown, undetermined, or unsuspected source. Sanitarians, laboratory analysts, and often an epidemiologist work together on investigations. Such a team may be assembled from different agencies and usually includes local officials, the FDA, and the Centers for Disease Control and Prevention (CDC) when the problem is large and widespread. The investigators will apply both standard procedures and special procedures necessitated by the particular case in determining which food caused the illness and/or how that food became contaminated. They will try to correlate their results with clinical information about the patients. From hard data and circumstantial evidence, they will proceed with reason, imagination, and caution to unravel the chain of events that caused contamination and illness. Detailed descriptions of two different outbreaks, one historical and the other rather recent, are given below to illustrate the characteristics of the investigation process.

YERSINIA ENTEROCOLITICA: FACT AND THEORY OF A FOODBORNE ILLNESS OUTBREAK

This incident involved milk that had been commercially pasteurized. The milk came from cows, but, another species of farm animal was also involved. The year was 1982, and in three south-central states of the United States—Tennessee, Arkansas, and Mississippi—up to 19,000 people became ill with diarrhea and other symptoms of clinical yersiniosis. Testing of 172 patients showed that the likely causative agent was the bacterium *Yersinia enterocolitica* of serogroup O:13 and typically the isolates carried a 42 megadalton plasmid.

The number of ill individuals was quite large, yet all seemed to be the customers of a single dairy. Milk from that supplier was the sole consumable item all these people had in common. That information surprised one of

the investigators on the team assembled to try to understand the situation. He was the FDA's Dr. Calvin Aulisio, and he knew that the O:13 serotype of *Y. enterocolitica* is associated endemically not with dairy cattle but, rather, with pigs. Another anomaly was that there appeared to be no contamination in the milk as sold by the dairy. All pasteurization records indicated that the required kill step for pathogens had been taken properly. Aulisio wanted to know if there was any association between the dairy and pigs. Interviewing practically everyone at the dairy from top officials down, he always received the same reply. No one knew of any contact, any connection between the dairy and pigs—until he came to the last person on his list: the employee on the dairy's loading dock. That man told Aulisio that outdated, unsold milk returned to the dairy was not destroyed but sold to a pig farm as feed for the pigs; however, there still seemed to be no contact between the pigs and the dairy, because all the milk and even the milk containers were left at the pig farm.

Aulisio decided to follow the dairy's delivery truck with the outdated milk to the pig farm. He did this several times and observed that although the pig pens were on a hill, the truck stopped at the bottom of the hill and that is where the crates containing the milk cartons and bottles were placed on the ground. Then the cartons and bottles were removed from the crates, which were placed back on the truck. In rainy weather, feces from the pig pens were washed down the hill and became mixed into the soil at the bottom of the hill where the truck parked and the crates were unloaded. When the reloaded empty crates reached the dairy again, they were washed with hot water; however, some soil remained in indentations in the outside bottoms of the crates. In other words, the washing was not thorough enough. Aulisio cultured soil from the bottom of the hill at the pig farm and soil remaining on the crates; both were positive for *Y. enterocolitica* serogroup O:13 carrying the 42 megadalton plasmid (Aulisio et al., 1982).

What likely happened? When containers of freshly pasteurized milk were placed in the incompletely washed crates and the crates were stacked on top of each other under refrigerated conditions, moisture accumulated and caused remnants of soil with the bacteria to drip down onto the milk containers below. Consumers either contaminated their hands when handling the milk containers and/or inoculated the milk inside when they opened the container. Pasteurized milk with its lack of a competitive microflora is a good growth medium for cold-tolerant *Yersinia* species. Experiments were conducted to determine whether the strain of *Yersinia* involved in the outbreak could survive on the outside of refrigerated milk containers (Stanfield et al., 1985). It survived well for as long as 21 days. So did some other foodborne pathogens. That gave support to the theory that the outside of food packaging can play a role in the transmission of infections. Lessons learned include that we must keep the outside of food containers clean and that persistence pays off in investigations.

SPRINGTIME FOR *CYCLOSPORA*: AN OUTBREAK STUDY WITH INTERNATIONAL ASPECTS

An illness with outbreaks in Canada and the United States during spring and early summer of 1996 had unusual features. Its symptoms were those of a strong, very long-lasting diarrhea. In addition, people described feeling "jet lag." The malady seemed to strike those who ate at upscale events—banquets, receptions, and country club gatherings. Identified in stool samples from the afflicted was a microorganism, presumably the cause of the illness that had been described and classified only 3 years earlier. It was a single-cell animal, a parasitic protozoa given the name *Cyclospora cayetanensis* (Ortega et al., 1993). The number of people who became ill with cyclosporiasis during the months of May, June, and very early July was 1465, about one-half of them having been to what one pundit termed "posh parties at plush places." Yet no cases occurred west of the Rocky Mountains. These circumstances caught the attention of the news media and stimulated the public's imagination.

It took time to determine what food at those expensive events had caused the illness. The list of possibilities came down to two fresh produce items: strawberries and raspberries; and it was the latter to which the data from interviews and questionnaires finally pointed (Herwaldt et al., 1997). The raspberries, it was then realized, had been imported and the country of their origin was Guatemala in Central America.

The raspberry is not native to Guatemala. It began to be cultivated there as a cash crop at the urging of international agencies and was intended mostly for export to Canada and the United States at those times of the year when there were gaps in the two countries' raspberry supply. Spring and autumn became the high points of Guatemalan raspberry production because the northern nations grew their own raspberries in summer and had been importing South American raspberries in winter. Guatemala's raspberry exports increased considerably from 1994 to 1996, the year the associated outbreaks of illness were so numerous they could not be overlooked.

To confirm the epidemiologic implication that Guatemalan raspberries were the carriers of cyclospora, scientists looked for the organism on the berries but could not find it. Several elution methods and detection techniques were tried, including direct observation by microscopy and genetic identification by the polymerase chain reaction. Detected by these means in the berry washes was not cyclospora but one of its relatives, another parasitic protozoa, an eimeria that is a parasite of birds and some mammals but does not cause human infections. This was a valuable finding. It showed that the analytical methods being used did work to some extent, but perhaps needed more refinement. It also showed that something from animals was getting onto the berries, and people wondered whether it was some animal that was contaminating the raspberries with cyclospora.

Government agencies in the United States and Canada were, however, expected to do more than develop finer detection methods and speculate about the source of infections. The next crop of raspberries from Guatemala would be arriving in the autumn of 1996. Should they be allowed entry?

It was suspected, but not definitely known, that the fall season was not the right one for cyclosporiasis. In Guatemala the illness was associated with the spring rainy season and was thought to be the cause or at least one of the causes of the country's long familiar springtime diarrheas.

Guatemalan raspberries were allowed into Canada and the United States in the autumn of 1996 and no associated illness outbreaks occurred. The real worry was the next springtime crop, in 1997. Guatemala was asked to inspect its raspberry fincas (farms) and only those classified as low risk for contamination would be allowed to export to Canada and the United States. The criteria used for the risk classification were not spelled out precisely. Again illnesses occurred. There were 762 cases in 41 clusters plus 250 sporadic cases that fit the definition of long-lasting diarrhea in April and May of 1997, a total of 1012 presumed cyclospora infections. They occurred in 17 states of the United States and in the District of Columbia, as well as in the same two Canadian provinces as the previous year. This time the spread was from the East Coast all the way to the West Coast and did not stop after the Rocky Mountains. Again the epidemiology pointed to fresh Guatemalan raspberries as the vehicle for the parasites, even though they were supposedly from low-risk farms.

In consultation the governments of Guatemala, Canada, and the United States decided to conduct risk analyses of the situation. The possible ways that berries became contaminated with cyclospora were several: (1) spraying with insecticide and fungicide dissolved in water that was not potable; (2) touching by the hands of the fieldworkers when being felt for ripeness and when being picked; (3) contamination through animal vectors; (4) exposure to physical forces (wind with dust, splatter from rain) in the fields; (5) accidental or intentional intermingling of berries from high-risk farms (i.e., unsanitary ones) with those from low-risk (i.e., sanitary farms); and (6) sabotage, the intentional contamination of berries with fecal matter that may have contained cyclospora.

Remedies were possible for some of the risks: (1) placing filters (1 μm) to exclude cyclospora (the infective oocyst has a diameter of 8-10 μm) in the spray apparatus for the insecticide and fungicide solutions; (2) monitoring fieldworkers' hand washing and general hygiene. Unfortunately, nothing effective could be done about risks 3 and 4, except deploying scarecrows to keep birds off the fields. Other risks were addressed by labeling berry containers to indicate the particular farm and day of harvest (risk 5); and security measures to discourage intermingling of produce from different farms and facilitate trace-backs in the event of illness outbreaks (risk 6). Risks 3 and 4 were considered much less likely to occur than the other risks. Remedies 1, 2, 5, and 6 together with farm

inspections and health checks of farm workers by an independent authority were implemented as a model plan of excellence for raspberry production in Guatemala. The Guatemalan government would assure importing nations that only those raspberries grown, packaged, and transported according to the plan would be exported.

Doing the risk analysis, implementing the remedies, and instituting the inspections took time. In 1998 much of the work had not been accomplished, and the United States decided not to accept any Guatemalan raspberries that spring. No cyclosporiasis outbreaks were detected in the United States that season. Canada did import fresh raspberries from Guatemala in the spring of 1998 and had 336 typical cases. Unintentionally, this amounted to a "controlled" epidemiological experiment. In 1999 the model plan of excellence (Jackson et al., 1999) for exporting raspberries was in full operation. The United States imported Guatemalan spring raspberries and experienced no cyclosporiasis outbreaks. Canada, careful due to its 1998 laxity, did not accept fresh Guatemalan spring raspberries again until 2002. The United States had no cyclosporiasis due to Guatemalan raspberries in 1999, 2001, 2002, or 2003. There were 63 cases in 2000, all traceable to a single farm, thanks to the labels on the raspberry containers. It is suspected that this farm experienced sabotage. The owner of the farm was found murdered.

Although it was demonstrated that contamination of raspberries with cyclospora could be prevented by following the plan, the total story of Guatemalan raspberry exports has a sad ending. The costs of implementing the plan, a sequence of years with too much rain for optimal raspberry production, and competition from other Latin American countries that presumably did not have endemic cyclosporiasis and so did not have to institute expensive preventive plans combined to make Guatemalan raspberry exports noncompetitive and not profitable. Still, there was a benefit. Guatemala has applied elements of the model plan of excellence to other farm crops, and these now have a high reputation for microbial safety on the world market.

There is a subplot to the story of investigating Guatemalan raspberries that concerns the detection methodology for *Cyclospora cayetanensis*. There is also a side story about Guatemalan blackberries and a sequel about raspberries from Chile.

Detection Methodology. Microscopy of raspberry washings took much time. Although eventually the *C. cayetanensis* oocyst was detected visually, as some *Eimeria* spp. oocysts had been in 1996 and 1997, the procedure was abandoned as impractical. Initial results with the polymerase chain reaction (PCR) were negative because of interfering substances, particularly when the raspberries were not fresh. Since these interfering substances could be removed by special filters (from Fraser Technology Australia), PCR became the method of choice (Lopez et al., 2001; Orlandi et al., 2004).

Why Raspberries and Not Blackberries? Raspberries and blackberries were grown in adjacent fields of the same Guatemalan farms during the same seasons. Why were just the raspberries implicated as vehicles for cyclosporiasis? (Actually, fresh Guatemalan springtime blackberries may have caused a small number of cyclosporiasis cases in Canada.) Blackberries have a smooth surface and are more washable than raspberries. Raspberries are covered by fine, sticky hairs to which all sorts of microscopic objects adhere: dust particles, pollen grains, and the cysts and oocysts of parasites.

Cyclosporiasis and Raspberries from Chile. In the winter of 2002 an outbreak in the United States of 22 cases of cyclosporiasis was traced to raspberries from Chile. It is thought that there is no endemic cyclosporiasis in Chile. How were the Chilean berries contaminated? Investigation (Schrimpf et al., 2003) suggested two possibilities: either by guest crop pickers from endemic countries, such as Peru, or because the infection is endemic in Chile but has not yet been detected owing to the limited number of surveys to date.

LESSONS LEARNED AND LESSONS TO BE LEARNED

Persistence again proved to be valuable in investigating the North American outbreaks of cyclosporiasis, tracing many of them to fresh raspberries (canning or freezing kill *C. cayetanensis*) and, by way of a risk analysis, suggesting interventions, such as the model plan of excellence, for the production and distribution of the berries. Other items of fresh produce that have caused some cyclosporiasis outbreaks in international trade are lettuce, basil, and as mentioned, blackberries. There is much yet to learn about cyclosporiasis. Why is it so seasonal? Where is it between seasons? Are there reservoir hosts or transfer hosts for the parasite? In how many countries and what environments is it endemic?

WHY ARE THERE SO MANY FOODBORNE INFECTIONS?

If one considers the passage of food from farmland or fishing waters to the consumer's fork, it is apparent that contamination control was for many years concentrated on food processing. The earlier steps of food growth and the later steps of distribution, sale, and consumer handling were not completely controlled or were not controlled at all. That situation improved somewhat in certain countries in the later 1990s. In the United States the improvement was spurred by the nation's Food Safety Initiative. Yet, the large size of food production firms and the wide distribution of their products magnifies incidents of contamination. Canning, which kills most pathogens, has been partially replaced by freezing, which may preserve the pathogen. Recycling, for economic purposes, of food waste back into the food chain also may recycle microbes. The use of antibiotics and preliminary heat in food production and processing

may be causing microbial resistance due to microbial adaptations and even mutations. The percentage of immunocompromised consumers has grown. More raw food is being eaten for dietary reasons in prosperous countries. As cities grow, particularly in poorer nations, there are more street vended foods and fewer supplies of potable water. Prevention advice is not always understood. For these and other reasons foodborne infections are still a problem in the 21st century.

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REFERENCES

Aulisio, C. C. G., M. T. Lanier, and M. A. Chappel. 1982. *Yersinia enterocolitica* O:13 associated with an outbreak in three southern states. Journal of Food Protection 45:1263.

Herwaldt, B. L., Ackers M. L., Cyclospora Working Group. 1997. An outbreak in 1996 of cyclosporiasis associated with imported raspberries. New England Journal of Medicine 336:1548-1556.

Jackson, G. J., L. A. Solorzano, J. E. Kvenberg, L. G. Flores, J. D. Mooney, and R. Castaneda. 1999. Routes by which *Cyclospora cayetanensis* may contaminate food. In Food microbiology and food safety into the next millennium. Proceedings, 17th Conference, International Committee on Food Microbiology & Hygiene, Veldhoven, Netherlands, eds. A. C. J. Tuijtelaars, R. A. Samson, F. M. Rombouts, and S. Notermans. Pp. 49-51.

Lopez A. S., D. R. Dodson, M. J. Arrowood, P. A. Orlandi Jr., A. J. da Silva, J. W. Bier, S. D. Hanauer, R. L. Kuster, S. Oltman, M. S. Baldwin, K. Y. Won, E. M. Nace, M. L. Eberhard, and B. L. Herwaldt. 2001. Outbreak of cyclosporiasis associated with basil in Missouri in 1999. Clinical Infectious Diseases 32:1010-1017.

Orlandi, P. A. Jr., and Fishman, P. H. 1998. Filipin-dependent inhibition of cholera toxin: Evidence for toxin internalization and activation through caveolae-like domains. Journal of Cell Biology 141:905-915.

Ortega, Y. R., C. R. Sterling, R. H. Gilman, V. A. Cama, and F. Diaz. 1993. Cyclospora species—A new protozoan pathogen of humans. New England Journal of Medicine 328:1308-1312.

Schrimpf, J. E., G. J. Jackson, J. J. Guzewich, I. Santiago, and T. Hill. 2003. FDA and Chile: Sharing science for produce safety. In 9th Annual Science Forum—FDA Science: Protecting America's Health; FDA, Rockville, Maryland, USA. P. 83.

Stanfield, J. T., G. J. Jackson, and C. C. G. Aulisio. 1985. *Yersinia enterocolitica*: Survival of a pathogenic strain on milk containers. Journal of Food Protection 48:947-948.