

A Multistate Outbreak of Cyclosporiasis

A Classroom Case Study

INSTRUCTOR'S VERSION

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NOTE: This case study is based on real-life investigations undertaken in 1996 and 1997 in the United States and abroad that were published in the Morbidity and Mortality Weekly Report, the New England Journal of Medicine, and the Annals of Internal Medicine. The case study, however, is not a factual accounting of the details from these investigations. Some aspects of the investigations (and the circumstances leading up to them) have been altered to assist in meeting the desired teaching objectives. Some details have been fabricated to provide continuity to the storyline.

Target audience: students with minimal knowledge of basic epidemiologic concepts who are interested in learning more about the practice of epidemiology including participants in the Knight Journalism Fellowship Program.

Level of case study: basic

Teaching materials required: none

Time required: approximately 3 hours

Language: English

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Learning objectives:

After completing this case study, the participant should be able to:

- 1) use the modes of transmission and incubation period for a disease to focus the search for the source of an outbreak
- 2) describe the two most common types of epidemiologic studies routinely used to investigate outbreaks
- 3) interpret the results of an epidemiologic study
- 4) consider potential sources of error in designing or carrying out an epidemiologic study
- 5) apply the criteria for causation to the results of an outbreak investigation
- 6) list considerations in implementing control measures before confirmation of the source of an outbreak
- 7) describe the occurrence, signs and symptoms, and control of cyclosporiasis

Part I – Background

On May 20, 1996, the following article appeared on the front page of the Toronto Sun:

<p>Exotic Parasite Sickens Canadian Businessmen</p> <p>By Xavier Onnasis</p> <p>TORONTO - Public health officials today confirmed that three Canadian businessmen, two from Toronto and one from Ottawa, were diagnosed with cyclosporiasis, a parasitic disease seen only in tropical countries and overseas travelers. The three men, who had recently traveled to the United States, became seriously ill with diarrhea over the weekend (May 16-18). One of the men was hospitalized at Princess Margaret Hospital when he collapsed due to severe dehydration.</p> <p>Dr. Richard Schabas, Ontario's Chief Medical Officer, reported that cyclosporiasis was exceedingly rare in North American and that much was still unknown about this disease. Cyclosporiasis is caused by the</p>	<p>microorganism <i>Cyclospora cayetanensis</i>. <i>Cyclospora</i> infects the small bowel and usually causes watery diarrhea, with frequent, sometimes explosive, bowel movements. Symptoms can include bloating, increased gas, stomach cramps, nausea, loss of appetite, and profound weight loss. The illness is diagnosed by examining stool specimens in the laboratory.</p> <p>Dr. Schabas declined to identify a source of infection for the three businessmen but indicated that the parasite is transmitted through contaminated food or water but not by direct person-to-person spread. The time between exposure to the parasite and becoming sick is usually about 7 days.</p> <p>Dr. Schabas reported that all three men had attended a meeting in Texas on May 9-10. He said Ontario Health Department staff would be investigating leads locally and in Texas.</p>
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See Appendix 1 for "Cyclosporiasis Fact Sheet".

Question 1: What is the incubation period for cyclosporiasis? How will it be used in the investigation?

*The incubation period is the **time between exposure to an infectious agent or toxin and the first appearance of symptoms** suggestive of the infection or intoxication. The incubation period of a disease can help investigators identify the agent causing an illness. For example, an incubation period of a few minutes or hours is suggestive of a preformed toxin (e.g., heavy metal, paralytic shellfish poisoning, *Staphylococcus aureus*); a few days, a viral or bacterial infection (e.g., norovirus, salmonellosis, *E. coli* O157:H7 infection), a week or more, a parasitic infection (e.g., cyclosporiasis, giardiasis). If the causative agent is known, the incubation period can help determine the most likely period of exposure to the agent (i.e., one incubation period before the onset of illness).*

*The **average** incubation period for cyclosporiasis is about 1 week but can range from 2-10 days.*

NOTE TO INSTRUCTORS: Students should be reminded of two good resources on the epidemiology of infectious diseases (including incubation period, modes of transmission, common sources of infection, and period of communicability):

- CDC Health Topics A to Z at <http://www.cdc.gov/az.do>
- Control of Communicable Diseases Manual published by American Public Health Association.

Question 2: On what sources of infection should public health officials focus for the three cases of cyclosporiasis? Is it possible that one of the men was the source of infection for the others? Do you think that it is likely that the businessmen became infected with cyclosporiasis in Texas?

*Cyclospora is spread by ingesting something that has been contaminated with stool containing the parasite (i.e., the **fecal-oral route**). Cyclospora **needs time** (days or weeks), however, after being passed in a bowel movement **to become infectious**. (The parasite, which is passed in the stool as an “oocyst,” must change forms and “sporulate” to become infectious.). As a result, food or water contaminated with unsporulated oocysts shortly before consumption should not cause infection and direct person-to-person spread is unlikely. In addition, exposure of the parasite to temperatures less than -20°C for 24 hours or above 60°C for 1 hour inactivates the oocysts. No documented outbreaks have been associated with cooked or commercially frozen foods.*

*In investigating the source of these cyclosporiasis cases, public health officials should focus on **foods, water, or beverages** that were consumed by the three men approximately **one incubation period** (i.e., about a week) **before becoming ill**. Officials may want to focus on uncooked foods and foods that have not been frozen (e.g., fresh produce) since cooking and freezing inactivate the parasite.*

Since all three men had onset of symptoms around the same time, it is unlikely that one was the source of infection for the others.

*The onset of symptoms among the three men was reported to be May 16-18. Counting back the average incubation period for cyclosporiasis (i.e., one week), the **period of exposure** for the three men was about May 9-11 (i.e., around the time they were in Texas). If they shared no other common exposures around that time period (e.g., meals on airlines, drinks at airport lounges), it is very likely that they became infected with cyclosporiasis in Texas.*

Part II – Outbreaks in Texas

The chief medical officer of the Ontario Health Department notified the Texas Department of Health (TDH) about the *Cyclospora* infections in the three Canadian businessmen. The businessmen had attended a meeting at a private club in Houston, Texas on May 9-10.

A total of 28 people had attended the Houston business meeting. Participants came from three U.S. states and Canada. Meals served during the meeting were prepared at the restaurant operated by the private club. Rumors among restaurant staff suggested that other attendees at the meeting had also become ill.

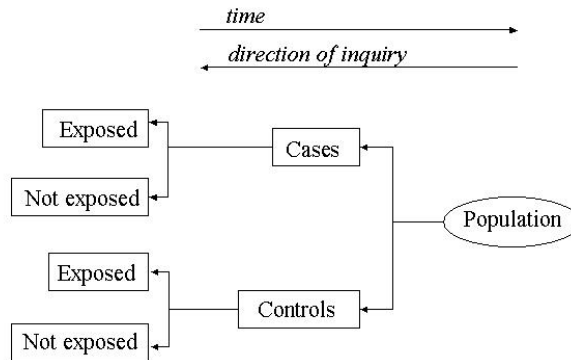
TDH, the Houston Health & Human Services Department, and the Centers of Disease Control and Prevention (CDC) initiated an epidemiologic investigation to identify the source of the cyclosporiasis outbreak.

Question 3: What are the two most common types of epidemiologic studies used to investigate the source of an outbreak (or other public health problem)? Which would you use to investigate the source of the cyclosporiasis outbreak in Texas? Why?

*In some outbreak investigations, laboratory, environmental, and/or epidemiologic information available at the outset so clearly supports a particular source for the outbreak that all that is needed is to carefully examine the established facts. In most investigations, however, the circumstances are not that straightforward and an **analytic study** (which includes a comparison group) must be undertaken. An analytic study allows investigators to quantify relationships between a suspected exposure and disease, test hypotheses about causal relationships, and explore the role of chance.*

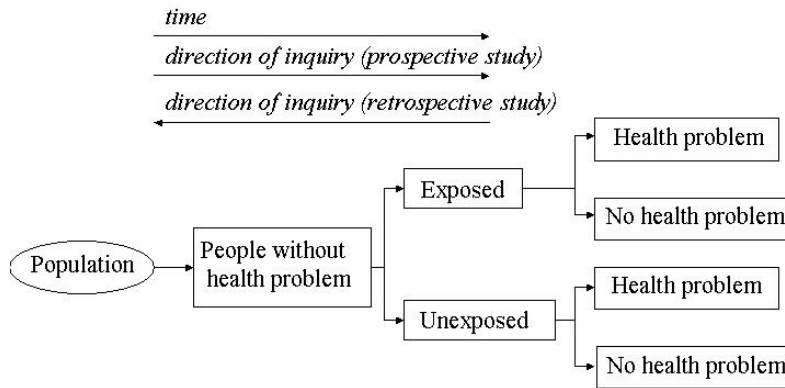
In outbreak investigations, two types of analytic studies are commonly used:

- **case-control study** – in a case-control study, subjects are enrolled based on whether they have (or had) the disease of interest (i.e., are a “case”) or not (i.e., are a “control”). Characteristics, such as previous exposure to some factor, are then compared between cases and controls to see if there is a relationship between the disease and the exposure.



from: Beaglehole, et al. (1993)

- cohort study** - in a cohort study, subjects are enrolled based on exposure (or lack of exposure) to a factor of interest (e.g., whether or not they smoked cigarettes, took a particular medication, ate a particular food) or if they were a member of a particular group (e.g., were a guest at a wedding banquet, were a patient on a particular hospital ward). The occurrence of disease is then ascertained and compared between people with exposure to the factor of interest and those without the exposure to see if there is a relationship between exposure and disease.



from: Beaglehole, et al. (1993)

When do you use which type of study? A case-control study is typically used when the population at risk is not known, when the disease under investigation is rare, when the exposure is common, or if the time between exposure and onset of the disease is long. A cohort study is typically used when illness occurs among a well-defined group or population, when exposure is rare, or when the disease is common.

The cyclosporiasis outbreak in Texas appears to be associated with a business meeting that involved a small, well-defined group of attendees. Therefore, it makes sense to undertake a **cohort study** to identify the source of the outbreak. Because the study was initiated after the outbreak occurred and will rely on the collection of exposure information after the fact (i.e., after disease has developed), the study is called a **retrospective cohort study**.

Because the outbreak appeared to affect a small, well-defined group of individuals (i.e., meeting attendees), investigators undertook a retrospective cohort study to investigate the source of the cyclosporiasis.

Investigators first surveyed people who attended the meeting to characterize the illness associated with the outbreak. (Twenty-seven of the 28 meeting attendees were interviewed.) All ill people experienced severe diarrhea and weight loss. In addition, 87% reported loss of appetite; 87% reported fatigue; 75% reported nausea; 75% reported stomach cramps; and 25% reported fever. Five ill people had stool specimens positive for *Cyclospora*.

Based on this information, investigators defined a case of cyclosporiasis for the cohort study as diarrhea of at least 3 days duration in someone who had attended the business meeting. Laboratory confirmation of *Cyclospora* infection was not required.

Of the 27 meeting attendees who were interviewed, 16 (59%) met the case definition for cyclosporiasis. Onsets of illness occurred from May 14 through May 19. (Figure 1)

Investigators questioned both ill and well meeting attendees about travel history and food and water exposures during the meeting.

Question 4: Why would you question people who did not become ill about possible sources of infection with *Cyclospora*?

Analysis of exposures among ill people (i.e., cases) alone is insufficient to determine the relationship between an exposure and a disease. Exposures common among cases may be related to the disease or may just be common in the group at large.

A comparison group is needed to determine how common exposure is in the group at large. *The comparison group provides a baseline for the level of exposure to the factor of interest (i.e., what you would expect to find in the group at large if the exposure were not associated with the disease). In the Texas cohort study, people who attended the Houston meeting and did not become ill served as the comparison group.*

Restaurant management at the private club refused to take calls from investigators or cooperate with the investigation. As a result, a list of foods served at meals during the meeting was obtained from the meeting organizer. No menu items were confirmed by restaurant staff.

Twenty-four meeting attendees provided information on foods eaten during the meeting. (Four attendees, including three cases, did not provide the information.) Investigators examined the occurrence of illness among people who ate different food items.

Twelve (92%) of 13 attendees who ate the berry dessert became ill. Only one (9%) of 11 attendees who did not eat the berry dessert became ill. The relative risk for eating berries was 10.2 (p-value <0.0001). No other exposures were associated with illness.

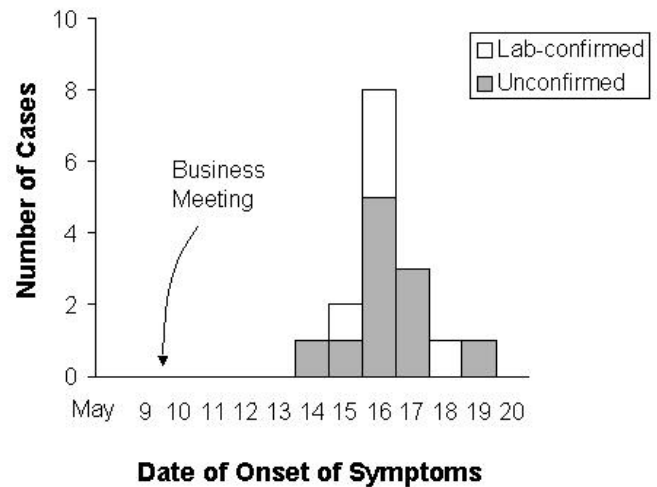
Case-patients reported that the berry dessert contained strawberries.

Question 5: In your own words, interpret the results of the cohort study.

The following statements can be made based on the results of the cohort study:

- *The attack rate for cyclosporiasis was **high among people who ate the berry dessert** (92%).*
- *The attack rate for cyclosporiasis was **low among people who did not eat the berry dessert** (9%).*
- *As a result, **the relative risk** (which compares the attack rate among people who ate the berry dessert with the attack rate among people who did not eat the berry dessert) **was high**. People who ate the berry dessert were over 10 times more likely to develop cyclosporiasis than those who did not.*
- ***Most** (12 out of 13) **of the cases of cyclosporiasis** (who provided food histories) **were exposed** to the berry dessert so that the exposure could “explain” almost all cases.*
- *There was a less than one-in-10,000 chance that the observed relative risk was due to chance alone.*

Figure 1. Onset of illness among patients with cyclosporiasis, Houston business meeting, May 1996.



Students interested in examining the calculation of the relative risk should be directed to Appendix 2.

Question 6: What problems in study design or execution should you consider when reviewing the results of this study (or any epidemiologic study)?

In reviewing the results of an epidemiologic study, one should always play devil's advocate and consider other possible explanations for the findings. The following items/questions should be routinely considered in interpreting any study results:

- **selection bias** (i.e., who got into the study) - Was participation in the study good? Were ill and well persons who participated in the study representative of all ill and well persons? (In the Texas study: Participation in the study was good with 24 [86%] of 28 meeting attendees providing food exposure information.)
- **information bias** (i.e., the validity of the information collected) - Was the information provided on exposure or disease development likely to reflect the truth? Did the exposures actually occur? Were cases really cases? Were individuals in the comparison group really well (i.e., could they have been asymptomatic or minimally symptomatic cases)? (In the Texas study: Only 5 [31%] of 16 cases were laboratory confirmed; however, the case definition [diarrhea lasting at least three days] reflected a fairly serious illness and probably identified true cases of cyclosporiasis. Since *Cyclospora* infection can be asymptomatic, some "well" persons may have actually been infected, but the proportion is unknown. Study participants had to recall what they ate a week earlier and may have had some difficulty providing accurate food histories from the meeting. In addition, it appears that food items served during the meeting were not confirmed with restaurant management.)
- **confounding** (i.e., other explanatory factors) - Is there another factor that is independently associated with both the exposure and the disease of interest that may distort the exposure-disease relationship? Is there an item consistently served with the implicated food that was actually the source of the infection? (In the Texas study: We do not know what analyses were performed to search for confounding.)
- **investigator error** - Were there problems with the execution of the study? Were enrollment criteria followed? Were case definitions consistently applied? Were data entry and calculations verified making investigator error remote? (In the Texas study: We do not know if there were problems in executing the study or with data entry or analysis.)

On June 4, before the first investigation had been completed, TDH was notified of another outbreak of cyclosporiasis involving physicians who attended a dinner on May 22 at a Houston, Texas restaurant. A second cohort study was undertaken. Nineteen attendees were interviewed. Ten met the case definition for cyclosporiasis (i.e., diarrhea of at least 3 days duration).

Attendees who ate dessert at the dinner were more likely to become ill than attendees who did not. Illness, however, was not associated with eating a particular type of dessert. No other exposures were associated with illness.

All desserts were garnished with either one fresh strawberry (for regular patrons) or with a strawberry, blackberry, and raspberry (for VIPs). Of the 7 attendees who reported eating a strawberry, all seven became ill. Of the eight attendees who reported not eating a strawberry, only one became ill (relative risk = 8.0, p-value = 0.001). (Note: four attendees, including two cases, could not recall whether they had eaten a strawberry and were excluded from this analysis.)

Based on the results of the two cohort studies, investigators hypothesized that strawberries were the source of the cyclosporiasis outbreaks in Houston.

Question 7: What additional studies might confirm (or refute) the hypothesis that strawberries were the source of the cyclosporiasis outbreaks?

Studies that might help confirm (or refute) the strawberry hypothesis include:

- *collection and testing of the implicated strawberries for Cyclospora*
- *traceback of the strawberries from the two events to the producer/distributor*
- *a search for additional cases of the disease (and their likely source)*

At the time of the outbreak, there was no effective testing procedure for Cyclospora on produce, so the likely utility of that action is questionable. If a traceback investigation of the strawberries from the two events found that the strawberries originated from the same producer/distributor or had specific places in common during production/distribution, that information would support the hypothesis that the strawberries were the source of the outbreak. Conversely, if the strawberries from the two events did not have any specific places in common, it would suggest that the strawberries were not the source of these outbreaks. Finally, because it is unlikely that the implicated strawberries were only distributed to the two restaurants in Houston associated with these outbreaks, other persons probably consumed them. And if the strawberries were the source of the outbreak, there would be other cases of cyclosporiasis in the community. A search for additional cases would be most beneficial in further exploring the source of the infection

TDH staff examined invoices and other records from the two restaurants involved in the Texas cyclosporiasis outbreaks. The strawberries consumed at both the May 9-10 business meeting and the May 22 physician dinner were grown in California. The individual producers/distributors of the strawberries, however, were not determined.

On May 31, TDH released a public health advisory about the presumed link between the consumption of California strawberries and the cyclosporiasis outbreak. The State Health Officer advised consumers to wash strawberries "very carefully" before eating them, and recommended that people with compromised immune systems (e.g., people with HIV infection, patients on cancer chemotherapy) avoid them entirely.

A few days later, Ontario's chief medical officer reported on an outbreak of cyclosporiasis in the Metro Toronto area affecting 40 people. Ontario public health officials believed California strawberries were also the source of the Toronto outbreak. A public health advisory, similar to the one from Texas, was issued.

Concurrent with the announcements from Texas and Ontario, CDC encouraged physicians from across the United States to report cases of cyclosporiasis to their local or state health department so that the source of the *Cyclospora* could be investigated further.

Question 8: You are writing a newspaper article about the cyclosporiasis outbreaks in Texas and Ontario. It is thought that the cyclosporiasis problem is ongoing. Four people are available for interview: the CDC expert on cyclosporiasis, one of the Canadian businessmen who became ill following the meeting in Houston, the owner of the private club in Houston where the first outbreak occurred, and the attorney for the California Strawberry Grower's Association. Your deadline is looming. You have time to ask each of these people only three questions. What would you ask them?

Since the New Jersey study focuses on laboratory-confirmed cases, review of laboratory records would be most appropriate.

Controls should be people without the health problem under investigation who are representative of the population from which the cases originated. They should be at risk for the health problem (i.e., they can develop the problem), have the potential for exposure to the factor(s) of interest, and be selected independent of their exposure to the factor(s) of interest. For the New Jersey study, investigators would want to use people with access to the same food sources as the cases. Therefore, they might use members of the case's family who are well or people living in the same neighborhood.

For the New Jersey case-control study, cases were identified by reviewing laboratory records from all clinical laboratories in the state. Forty-two cases were identified. Two controls were identified for each case through telephone calls to randomly selected households in the community. To be eligible for the study, controls could not have had loose stools during the previous 30 days.

Investigators interviewed 30 case-patients and 60 controls by telephone using a standardized questionnaire that asked about possible exposures (including consumption of 17 fruits and 15 vegetables, water and soil exposures, and animal contact) during the period of interest.

Case-patients and controls were similar with respect to age, sex, and level of education. Twenty-one (70%) of 30 case-patients and four (7%) of 60 controls had eaten raspberries. The odds ratio for eating raspberries was 32.7 (p-value <0.00001). No other exposures (including strawberries) were associated with illness.

Question 11: In your own words, interpret the results of the New Jersey case-control study.

The odds of exposure to raspberries was over 32 times higher among cases of cyclosporiasis than among controls. The probability that this finding was due to chance alone was less than one-in-a-million (i.e., the finding was statistically significant). In addition, this exposure could explain 70% of cases.

If investigators can rule out selection bias, information bias, confounding, and investigator error, the findings suggest that eating raspberries was a risk factor for cyclosporiasis in New Jersey.

Studies from other states and Canada supported the results from New Jersey, New York City, and South Carolina. A total of 725 cases of cyclosporiasis associated with 55 different events (e.g., wedding receptions, parties, buffets) were investigated. The only exposure consistently associated with cyclosporiasis was the consumption of raspberries.

Raspberries were served at 54 of the 55 events and were the only berries served at 11 events. (Reexamination of the events associated with the initial outbreaks in Texas and Ontario indicated that raspberries were included among the implicated berry items served at those events.) The median attack rate for cyclosporiasis among persons who ate items that contained raspberries at the different events was 93%. Furthermore, for 27 of the 41 events for which adequate data were available, the associations between the consumption of raspberries and cyclosporiasis were statistically significant (p-value<0.05).

The origin (i.e., producer) and mode of contamination of the raspberries served at the events were unknown. Due to the large number of raspberry producers at the time of the outbreaks (both domestic and international), public health officials could not recall the implicated raspberries or remove them from the marketplace. Traceback investigations were planned.

Question 12: Would you alert the public of this possible public health threat? Defend your answer.

NOTE TO INSTRUCTORS: Have students vote on this question. Then select a few voting for and against to defend their opinions. (There is no right or wrong answer. It is largely a matter of judgment.)

Public health officials often struggle with decisions similar to this one (called the “Grey Zone” in the business). Going public at this point means that you will tell people about a problem without being able to tell them what to do about it. And, broad/vague warnings can unnecessarily harm products not responsible for the problem and undermine the public’s confidence in the public health system.

Pros

Alerting the public to the linkage between raspberries and cyclosporiasis will result in consumers avoiding raspberries, in general. If the exposure is ongoing (i.e., contaminated raspberries are still available), this will prevent additional cases of cyclosporiasis from occurring as well as the morbidity, mortality, and costs associated with those cases. Avoidance of infection would be particularly critical for individuals at risk for serious complications of the illness (e.g., children under five years of age, the elderly, immunocompromised individuals).

Cons

Without specification of the producer, consumers will avoid all raspberries, resulting in an economic hardship for the entire raspberry industry. Consumers may continue to avoid raspberries even after the specific source of the problem is identified and corrected, taking months for individual producers and the industry to recover. In addition, vague or nonspecific warnings will heighten the public’s anxiety but not provide them with a means to do anything about it. If warnings continue to be non-constructive, the public will eventually stop listening to them.

Part IV – Traceback and Environmental Investigations

To identify the sources of raspberries served at the 54 events linked to outbreaks of cyclosporiasis, CDC, the U.S. Food and Drug Administration (FDA), and health departments from the affected states obtained information on the place and dates of purchase of the implicated raspberries. Distributors and importers of the raspberries were identified through invoices and shipping documents. Airway bill numbers and importation documents (e.g., Custom's forms), supplied by importers, were used to identify overseas shipments and exporters.

By the third week of July, investigators had documented the source of the raspberries for 29 of the 54 cyclosporiasis-associated events. For 21 of these events, the raspberries definitely came from Guatemala. For 8 events, the raspberries could have originated there. No commonalities were found in the U.S. ports of entry for the implicated raspberries.

During the outbreak period, raspberries had been imported from a number of countries. Based on monthly data from the U.S. Department of Agriculture, Guatemalan raspberries represented 4-20% of fresh raspberries (domestic and imported) shipped within the United States in April-June of 1996.

Question 13: Does the traceback information support raspberries as the source of the cyclosporiasis outbreak?

The fact that a large proportion of the raspberries associated with the outbreaks were linked to a common place (i.e., Guatemala) is strong evidence that they were the source of the outbreak. This finding is even more notable given that only 4-20% of raspberries shipped within the United States during that time period came from Guatemala.

NOTE TO INSTRUCTORS: The implicated food item was fresh produce which is notoriously difficult to trace due to the short shelf-life, multiple possible shipments, and lack of brand-name recognition. Although glossed over in the above description, much time and effort went into the documentation of the source of the raspberries.

At the time of the investigation, seven Guatemalan exporters, of which A and B were the largest, shipped raspberries to the United States. The raspberries for 25 of the 29 events were traced to only one Guatemalan exporter per event:

- 18 of 25 (72%) to Exporter A
- 5 (20%) to Exporter B
- 1 (4%) to Exporter C
- 1 (4%) to Exporter D

Using exporter records, the raspberries were traced back to the farm where they were grown. Because exporters typically combined raspberries from multiple farms in a shipment, investigators could identify only a group of contributing farms (an average of 10 farms with a range of 2 to 30) rather than one source farm per event. More than 50 farms could have contributed to implicated shipments of raspberries.

To investigate how raspberries were grown and handled in Guatemala, CDC and FDA investigators visited Exporters A, B, C, and D and the seven most commonly implicated raspberry producing farms (six supplying Exporter A and one supplying Exporter B).

Question 14: Given what you know about the transmission of cyclosporiasis, on what cultivation or harvesting practices would you focus in the investigation of the raspberry-producing farms?

One would want to focus on activities that could allow the raspberries to become contaminated with feces containing Cyclospora. This would include:

- *irrigation water (and source wells)*
- *proximity of fields or irrigation water to sources of human fecal material (e.g., pit latrines, outhouses, privies, and septic systems)*
- *substances added to the soil which could contaminate the raspberries (e.g., manure, fertilizer)*
- *substances applied to the raspberries (e.g., insecticides, fungicides, and liquids used to dilute these substances)*
- *harvesting or processing equipment or surfaces that might come in contact with the raspberries*
- *human handling of raspberries*
- *storage containers*
- *shipping trucks*

NOTE: The focus of the investigation was on contamination of raspberries with human feces. However, it was not and is still not known if animals can become infected with Cyclospora or be a source of infection for humans. Therefore, this focus may not have been appropriate.

The six most commonly implicated farms supplying Exporter A were in the same region of Guatemala. All six began harvesting for the first time in 1996 and often had raspberries in the same shipment. Five of the farms obtained agricultural water from wells. These wells varied in construction, depth, and quality. Two of the five farms also stored well water in reservoirs constructed of concrete blocks and covered with concrete. The sixth farm used river water. The farm that supplied raspberries to Exporter B was 25 km from the closest of the six farms that sold raspberries to Exporter A. That farm used well water, which was stored in a mesh-covered, plastic-lined, man-made reservoir.

At all seven farms, ground-level drip irrigation was used (primarily during the dry season) to avoid direct contact between raspberries and water. Agricultural water was also used to mix insecticides and fungicides that were sprayed directly onto raspberries, sometimes as late as the day they were harvested. At all farms, the raspberries were picked and sorted by hand, packed in plastic containers, and flown to the United States within 36 hours of picking.

Agricultural water at the seven farms (and on Guatemalan raspberry farms, in general) was filtered to remove debris but not microbes. Testing of agricultural-water samples from the seven farms indicated at least intermittent contamination with bacteria commonly found in sewage and human wastes (i.e., “coliforms” such as *Escherichia coli*). No *Cyclospora* were found.

No *Cyclospora* were found in samples of Guatemalan raspberries obtained from the farms during the traceback investigation.

Question 15: *Cyclospora* were not found in any Guatemalan raspberries or water samples. If the Guatemalan raspberries were the source of the cyclosporiasis outbreaks, list plausible explanations for this finding.

There are several possible explanations for not finding Cyclospora on Guatemalan raspberries or water samples at the time of the traceback investigations:

- ***Testing methods** for Cyclospora were **insensitive**. A small number of Cyclospora organisms is probably enough to make people sick but the available methods for testing food for Cyclospora were not adequately sensitive to detect low levels of contamination. At the time of the outbreak, Cyclospora had never been isolated from any food item or water.*
- *The water and raspberries from the farms were **collected after the outbreak period**. Leftover raspberries from the cyclosporiasis-associated events in the United States and Canada were not available for testing. If contamination was temporary or intermittent (e.g., associated with environmental conditions such as excessive rain and flooding), one might not expect to find contaminated raspberries or water at the time of the traceback investigations.*
- *The contamination of the raspberries could have occurred **during transport and/or shipping**.*

Of course, some might interpret this finding to mean that the raspberries were not the source of the outbreak of cyclosporiasis. (But this is an unlikely explanation given the rest of the evidence!)

Investigators hypothesized that the raspberries became contaminated through spraying with insecticides and fungicides that had been mixed with contaminated water from improperly constructed or maintained wells near deep pit latrines or seepage pits. The wells may have been particularly vulnerable to contamination during the rainy season (e.g., from surface-water runoff), when the 1996 outbreak occurred. Once contaminated, the raspberries remained contaminated until eaten because they were too fragile and covered with crevices to be washed thoroughly.

By July 18, 1996, CDC and FDA declared that raspberries from Guatemala were the likely source of the *Cyclospora* outbreak.

Part V – Control and Prevention Measures

Although skeptical of study findings and suspicious of potential trade barriers, the government of Guatemala, Guatemalan raspberry growers and exporters, and the Guatemalan Berries Commission (a growers' organization) collaborated with CDC and FDA to decrease the risk of contamination of Guatemalan raspberries during growth, harvest, and packaging.

Question 16: What specific measures would you suggest to decrease the likelihood of contamination of raspberries from the Guatemalan farms?

The following measures will help decrease the likelihood of contamination of raspberries with Cyclospora at the Guatemalan farms:

- *Renovate (or construct new) wells and reservoirs so that potable water supplies are consistently safe for drinking and not vulnerable to contamination during periods of excessive surface-water runoff.*
- *Use potable water for drinking, handwashing, cleaning surfaces that touch berries, and mixing substances sprayed onto berries (e.g., insecticides, fungicides).*
- *Instruct workers on personal hygiene and the need to wash hands after going to the bathroom and before handling the raspberries or touching surfaces that will come into contact with the berries.*
- *Provide adequate sanitary and handwashing facilities for workers to decrease the potential for direct or indirect contamination of berries.*
- *Work with applied food safety researchers to identify methods to: 1) decrease cross-contamination of berries on surfaces used to sort and inspect them and 2) inactivate Cyclospora oocysts on raspberries using a treatment that does not affect the quality or taste of the berries (e.g., gamma irradiation).*
- *If contaminated raspberries are identified, undertake traceback investigations to identify the source farm. To facilitate these traceback investigations in the future, distributors should indicate the source farm on all containers (e.g., clamshell containers) used to package the raspberries for sale and invoices.*

The Guatemalan raspberry growers voluntarily improved employee hygiene, sanitation, and water quality. They implemented systems to monitor the production of the raspberries so that potential points of contamination could be identified and addressed (i.e., Hazard Analysis and Critical Control Point systems). The Guatemalan Berries Commission established a farm classification system (with only farms in the best class permitted to export) in an attempt to minimize the exportation of *Cyclospora*-contaminated raspberries to the United States.

During the fall and winter of 1996, no outbreaks of cyclosporiasis in the United States were linked to Guatemalan raspberries. In the spring of 1997, however, another multistate outbreak occurred. By the end of May, more than a thousand new cases of cyclosporiasis had been reported from 18 states and two provinces in Canada. Consumption of raspberries was strongly associated with the outbreaks and the preponderance of the traceback data implicated Guatemala as the source of the raspberries, suggesting either some farms did not fully implement the control measures or the contamination was associated with a source against which these measures were not directed.

In the face of warnings by U.S. public health authorities on the danger of eating Guatemalan raspberries, the government of Guatemala and the Guatemalan Berries Commission voluntarily suspended exports of fresh raspberries to the United States on May 30, 1997. (See "FDA Talk Paper".) The interruption of exportation caused large economic losses for the producers, especially small and medium-sized ones.

FDA

TALK PAPER

*Food and Drug Administration
U.S. Department of Health and Human Services
Public Health Service 5600 Fishers Lane Rockville, MD 20857*

T97-22

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OUTBREAKS OF CYCLOSPORIASIS AND GUATEMALAN RASPBERRIES

We have been receiving inquiries about recent U.S. outbreaks of cyclosporiasis, a diarrheal illness resulting from ingestion of the *Cyclospora* parasite. The following may be useful for answering questions.

According to the U.S. Centers for Disease Control and Prevention, fresh raspberries from Guatemala are the likely cause of outbreaks of cyclosporiasis that have occurred since mid-April in at least seven states including California, Maryland, Nebraska, Nevada, New York, Rhode Island and Texas. CDC and FDA are cooperating in investigating the outbreaks, examining epidemiological evidence and tracing the source of implicated raspberries.

FDA and CDC were informed by the Guatemalan government and the Guatemalan Berries Commission on May 30 that the country's growers voluntarily suspended shipment of fresh raspberries to the United States. FDA is working with CDC, the Guatemalan government and the Guatemalan Berries Commission to determine when the country may resume shipment of fresh raspberries to the United States.

FDA believes that few if any fresh raspberries from Guatemala remain on the U.S. market due to the short shelf life of the commodity. However, if any consumers, distributors, caterers, restaurateurs or retail establishments are holding fresh raspberries labeled as Product of Guatemala, they should not serve, sell or eat them.

FDA is advising consumers who have recently eaten fresh raspberries from Guatemala and who are suffering diarrheal illness to see a doctor for diagnosis and possible treatment for cyclosporiasis.

Cyclospora infects the small intestine and causes watery diarrhea with frequent, sometimes explosive bowel movements. Other symptoms include loss of appetite, substantial loss of weight, bloating, increased gas, stomach cramps, nausea, vomiting, muscle aches, low-grade fever and fatigue. Symptoms usually develop about a week after consuming contaminated product. *Cyclospora* infection can be successfully treated with appropriate antibiotics.

FDA will convene a public meeting in July to review the science on *Cyclospora* on fresh produce and its control. The date and other details of that meeting will be made public when they are available.

####

CDC and FDA continued to work with the government of Guatemala and the Berries Commission to determine when the safety of Guatemalan raspberries could be assured and exports could resume. The exportation of raspberries resumed in mid-June; however, U.S. public health authorities continued to warn of the dangers of eating Guatemalan raspberries.

In December 1997, amid objections from the Guatemalan government, FDA announced that it was blocking imports of raspberries from Guatemala for 1998. Before this time, FDA rarely denied imports without physical evidence, and this ban was based only on epidemiological evidence about past outbreaks and FDA observations on current raspberry production practices. Congressional representatives and supporters of free trade railed about protectionism and questioned the science behind the decision since *Cyclospora* had not been identified on any raspberries from Guatemala.

Question 17: Do you believe that the raspberries were the source of the multistate outbreaks of cyclosporiasis? Which of the criteria for causality (i.e., strength of association, biological plausibility, consistency with other studies, exposure precedes disease, and dose-response effect) have been satisfied in the linkage between raspberries and cyclosporiasis? How would you convince others on the validity of these findings?

Although no smoking gun was found (i.e., raspberries contaminated with Cyclospora), there is good epidemiologic, laboratory, and environmental evidence that the raspberries were the source of the outbreak:

- **strength of association** - *The median attack rate at the cyclosporiasis-associated events in North America was high among persons who ate items that contained raspberries. A statistically significant association between raspberries and cyclosporiasis was determined for a large proportion of the investigations of these events. Data from case-control studies of sporadic cases of cyclosporiasis in New York City, New Jersey, and Florida also demonstrated strong associations between illness and raspberries (large odds ratios).*
- **biological plausibility** - *The possibility of foodborne transmission of Cyclospora had been considered in previous outbreaks including an outbreak that occurred in Florida in 1995. Environmental investigations identified several potential sources of contamination during growth, harvest, and packaging of the raspberries. Water supplies at a number of the Guatemalan farms (used for irrigation and dilution of insecticides and fungicides) were found to be contaminated with E. coli, and could have contained other contaminants from human wastes. The timing of the outbreaks occurred during the rainy season when surface-water runoff would have been at its greatest, increasing the risk of contamination of suboptimally constructed or maintained wells near deep pit latrines or seepage pits.*
- **consistency with other studies** - *Multiple investigations (by different investigators) identified raspberries as the source of the cyclosporiasis outbreaks associated with events in North America. Traceback investigations of raspberries served at these events repeatedly led to Guatemalan raspberries. The timing of cases coincided with rising and falling imports of raspberries from Guatemala and the outbreaks ended after suspension of raspberry exports from Guatemala.*
- **exposure precedes disease** - *Exposure to raspberries preceded illness in both event-associated and sporadic cases in North America. The median incubation period for cases associated with events in North America was 7 days and ranged from 1 to 14 days (by definition, <15).*
- **dose-response effect** - *No information/data supporting this criterion for causation was reported in the literature.*

The U.S. ban on importation of Guatemalan raspberries became effective on March 15, 1998 and continued through August 15, the normal Guatemalan raspberry exporting season. With the ban in place, outbreaks of cyclosporiasis did not occur in 1998 in the United States.

Canadian officials decided not to block the importation of Guatemalan raspberries in 1998. In May and June, a multicluster outbreak of cyclosporiasis occurred in Ontario involving over 300 people. Investigations linked the outbreak to raspberries from Guatemala.

Beginning in the spring of 1999, the United States allowed entry of raspberries from farms that complied with a detailed program of food safety practices and successfully passed Guatemalan government inspections and FDA audits. That spring, there were several cyclosporiasis outbreaks in the United States and Canada; however, Guatemalan raspberries were not implicated as a source for any. In 2000, two outbreaks of cyclosporiasis were linked to raspberries traced to one Guatemalan farm. That farm discontinued exportation of raspberries.

As of June 2004, no further outbreaks of cyclosporiasis have been associated with Guatemalan raspberries. However, only three of the original 85 Guatemalan raspberry growers continue to export raspberries.

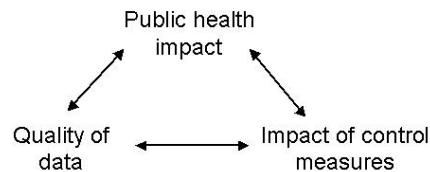
Epilogue

Announcements by Texas and Ontario public health officials implicating California strawberries as the source of the cyclosporiasis outbreaks in May of 1996 had a devastating effect on the strawberry industry. Supermarket chains took California strawberries off their shelves, in response to pressure from consumers. Consumers stopped buying strawberries from all sources. Truckloads of strawberries headed for market rotted as they were turned away by produce and grocery store managers. Strawberry sales around the United States and Canada crashed, causing \$40 million in losses for the industry and the loss of 5,000 jobs.

And, in the end, the actual vehicle for the outbreak turned out to be Guatemalan raspberries, not strawberries.

Question 18: To prevent additional cases of a health problem (and possible hospitalizations and deaths), public health authorities are often required to make decisions on control measures when data are suggestive of the source of the problem but are, perhaps, not conclusive. What criteria would cause you to implement control measures for a health problem before you were absolutely certain of the source?

*In making timely decisions to implement control measures, public health officials must **balance** the potential public health impact of a problem with the known quality of available data and the potential damage to business or industry.*



Information that might lead officials into taking action when data are suggestive of the source but insufficient to make a definitive call include:

- **the severity of the disease** (e.g., *E. coli* O157:H7, botulism) (i.e., one may be moved to act more quickly with a very serious or potentially fatal disease than one which is mild or self-limiting)
- **the population at risk** (e.g., infants, immunocompromised persons, the elderly) (i.e., if the population at risk includes persons who are highly likely to have poor outcomes from the infection/intoxication, then one may be moved to act more quickly)
- **whether exposure is suspected to still be occurring**
- **the quality of available data** (e.g., use of a controlled study of sufficient size to detect differences, low probability of selection or information bias or confounding, consistency with other studies)
- **potential impact on business/industry** (e.g., is the decision likely to impact an entire industry or a single business or foodhandler?)

*So what went wrong in Texas? In actuality, public health officials had data that **properly** implicated “berry dessert items” served at the two Houston events linked with the cyclosporiasis outbreaks. However, their results could not distinguish between the ingredients in the dessert and reports were inconsistent concerning the types of berries in the dessert. Based on information from case-patients from the Houston business meeting (who recalled that the dessert contained strawberries) and under pressure to act quickly, Texas investigators declared that the source of the outbreak was strawberries from California. If Texas investigators had considered results from other investigations occurring at the same time (New York City and South Carolina) or completed tracebacks of the California strawberries (which probably would not have led to the same producers/distributors), they may have been able to clarify the potential risk from the strawberries, raspberries, or other ingredients in the desserts.*

References

- CDC. Outbreaks of *Cyclospora cayetanensis* infection -- United States, 1996. *Morbidity and Mortality Weekly Report* 1996;45(25):549-551.
- CDC. Update: Outbreaks of *Cyclospora cayetanensis* infection -- United States and Canada, 1996. *Morbidity and Mortality Weekly Report* 1996;45(28):611-612.
- CDC. Outbreaks of cyclosporiasis -- United States, 1997. *Morbidity and Mortality Weekly Report* 1997;46(20):451-452.
- CDC. Update: Outbreaks of cyclosporiasis -- United States, 1997. *Morbidity and Mortality Weekly Report* 1997;46(21):461-462.
- CDC. Update: Outbreaks of cyclosporiasis -- United States and Canada, 1997. *Morbidity and Mortality Weekly Report* 1997;46(23):521-523.
- Herwaldt BL, Ackers ML. An outbreak in 1996 of cyclosporiasis associated with imported raspberries. The Cyclospora Working Group. *N Engl J Med* 1997;336:1548-56.
- Herwaldt BL, Beach MJ. The return of *Cyclospora* in 1997: Another outbreak of cyclosporiasis in North America associated with imported raspberries. *Annals of Internal Medicine* 1999;130:210-220.
- Herwaldt BL. *Cyclospora cayetanensis*: A review, focusing on the outbreaks of cyclosporiasis in the 1990s. *Clin Infect Dis* 2000;31:1040-57.
- Texas Department of Health. Outbreaks of *Cyclospora* infection. *Disease Prevention News* 1997;57(11):1-4.

APPENDIX 1: CDC Cyclosporiasis Fact Sheet (available at <http://www.cdc.gov/az.do>)

Cyclospora cayetanensis (SIGH-clo-SPORE-uh KYE-uh-tuh-NEN-sis) is a parasite composed of one cell, too small to be seen without a microscope. The first known human cases of illness caused by *Cyclospora* infection (i.e., cyclosporiasis) were reported in 1979. Cases began being reported more often in the mid-1980s. In the last several years, outbreaks of cyclosporiasis have been reported in the United States and Canada.

How is *Cyclospora* spread?

Cyclospora is spread by people ingesting something, for example, water or food that was contaminated with infected stool. For example, outbreaks of cyclosporiasis have been linked to various types of fresh produce. *Cyclospora* needs time (days or weeks) after being passed in a bowel movement to become infectious. Therefore, it is unlikely that *Cyclospora* is passed directly from one person to another. It is unknown whether animals can be infected and pass infection to people.

Who is at risk for infection?

People of all ages are at risk for infection. In the past, *Cyclospora* infection was usually found in people who lived or traveled in developing countries. However, people can be infected worldwide, including the United States.

What are the symptoms of infection?

Cyclospora infects the small intestine (bowel) and usually causes watery diarrhea, with frequent, sometimes explosive, bowel movements. Other symptoms can include loss of appetite, substantial loss of weight, bloating, increased gas, stomach cramps, nausea, vomiting, muscle aches, low-grade fever, and fatigue. Some people who are infected with *Cyclospora* do not have any symptoms.

How soon after infection will symptoms begin?

The time between becoming infected and becoming sick is usually about 1 week.

How long will symptoms last?

If not treated, the illness may last from a few days to a month or longer. Symptoms may seem to go away and then return one or more times (relapse).

What should I do if I think I may be infected?

See your health care provider.

How is *Cyclospora* infection diagnosed?

Your health care provider will ask you to submit stool specimens to see if you are infected. Because testing for *Cyclospora* infection can be difficult, you may be asked to submit several stool specimens over several days. Identification of this parasite in stool requires special laboratory tests that are not routinely done. Therefore, your health care provider should specifically request testing for *Cyclospora*. Your health care provider may have your stool checked for other organisms that can cause similar symptoms.

How is infection treated?

The recommended treatment for infection with *Cyclospora* is a combination of two antibiotics, trimethoprim-sulfamethoxazole, also known as Bactrim*, Septra*, or Cotrim*. People who have diarrhea should rest and drink plenty of fluids.

I am allergic to sulfa drugs; is there another drug I can take?

No alternative drugs have been identified yet for people who are unable to take sulfa drugs. See your health care provider for other treatment recommendations.

How is infection prevented?

Avoiding water or food that may be contaminated with stool may help prevent *Cyclospora* infection. People who have previously been infected with *Cyclospora* can become infected again.

For more information:

1. CDC. Outbreak of cyclosporiasis -- northern Virginia-Washington, D.C.-Baltimore, Maryland, metropolitan area, 1997. *MMWR* 1997; 46:689-91.
2. Herwaldt BL, et al. An outbreak in 1996 of cyclosporiasis associated with imported raspberries. *N Engl J Med* 1997;336:1548-56.
3. Herwaldt BL, et al. The return of *Cyclospora* in 1997: another outbreak of cyclosporiasis in North America associated with imported raspberries. *Ann Intern Med* 1999;130:210-20.
4. Hoge CW, et al. Placebo-controlled trial of co-trimoxazole for cyclospora infections among travellers and foreign residents in Nepal. *Lancet* 1995;345:691-3.
5. Hoge CW, et al. Epidemiology of diarrhoeal illness associated with coccidian-like organism among travellers and foreign residents in Nepal. *Lancet* 1993;341:1175-9.
6. Huang P, et al. The first reported outbreak of diarrheal illness associated with *Cyclospora* in the United States. *Ann Intern Med* 1995;123:409-14.
7. Ortega YR, et al. Cyclospora species - a new protozoan pathogen of humans. *N Engl J Med* 1993;328:1308-12.
8. Soave R. *Cyclospora*: an overview. *Clin Infect Dis* 1996;23:429-37.
9. Soave R, et al. Cyclospora. *Infect Dis Clinics N Amer* 1998;12:1-12.

APPENDIX 2: Calculating Measures of Association

Cohort studies

The **relative risk** is the measure of association for a cohort study. It tells us how much more likely (or less likely) it is for people exposed to a factor to develop a disease compared to people not exposed to the factor.

The relative risk is the **ratio of the attack rates** of a disease among people exposed to the factor and those not exposed to that factor. (The **attack rate** is the incidence of disease in a group [i.e., the number of people in the group who became ill divided by the total number of people in the group].)

$$\begin{aligned} \text{attack rate} &= \frac{\text{\# ill people in group}}{\text{\# people in group}} \\ \text{relative risk} &= \frac{\text{attack rate for exposed persons}}{\text{attack rate for unexposed person}} \end{aligned}$$

A relative risk of:

- **1.0** (or close to 1.0) means the risk of disease is similar in the exposed and unexposed group and exposure is not associated with disease.
- **Greater than 1.0** means the risk of disease is greater in the exposed than the unexposed group and the exposure could be a risk factor for the disease.
- **Less than 1.0** means the risk of disease is less in the exposed group than the unexposed group and the exposure could be a protective factor.

In the Texas cohort study:

Twelve of 13 attendees who ate the berry dessert became ill. Only one of eleven attendees who did not eat the berry dessert became ill. Inserting these numbers into the 2x2 table:

Ate Berry Dessert	Ill	Well	TOTAL
Yes	12	1	13
No	1	10	11
TOTAL	13	11	24

$$\text{attack rate (ate berries)} = \frac{\text{\# people who ate berries who became ill}}{\text{\# people who ate berries}}$$

$$= \frac{12}{13} = 92\%$$

$$\text{attack rate (did not eat berries)} = \frac{\text{\# people who did not eat berries who became ill}}{\text{\# people who did not eat berries}}$$

$$= \frac{1}{11} = 9\%$$

$$\text{relative risk (eating berries)} = \frac{\text{attack rate for people exposed to berries}}{\text{attack rate for people not exposed to berries}}$$

$$= \frac{92\%}{9\%} = 10.2$$

Interpretation: People exposed to the berry dessert were 10 times more likely to develop illness than people not exposed to the berry dessert.

Case-control studies

The **odds ratio** is the measure of association for a case-control study. It tells us how much higher the odds of exposure is among cases of a disease compared with controls.

The odds ratio compares the **odds of exposure** to the factor of interest **among cases** to the odds of exposure to the factor **among controls**. (The **odds** is the probability that an event will happen divided by the probability that it won't happen.)

For an unmatched case-control study, the data look like this:

Exposed	Case	Control	TOTAL
Yes	a	b	a+b
No	c	d	c+d
TOTAL	a+c	b+d	a+b+c+d

$$\begin{aligned} \text{odds of exposure (cases)} &= \frac{\text{number of cases with the exposure}}{\text{number of cases without the exposure}} = \frac{a}{c} \\ \text{odds of exposure (controls)} &= \frac{\text{number of controls with the exposure}}{\text{number of controls without the exposure}} = \frac{b}{d} \\ \text{odds ratio} &= \frac{\text{odds of exposure (cases)}}{\text{odds of exposure (controls)}} = \frac{ad}{bc} \end{aligned}$$

An odds ratio of:

- **1.0 (or close to 1.0)** means that the odds of exposure among cases is the same as the odds of exposure among controls. The exposure is not associated with the disease.
- **Greater than 1.0** means that the odds of exposure among cases is greater than the odds of exposure among controls. The exposure may be a risk factor for the disease.
- **Less than 1.0** means that the odds of exposure among cases is lower than the odds of exposure among controls. The exposure may be protective against the disease.

In the New Jersey case-control study:

Twenty-one of 30 case-patients and four of 60 controls had eaten raspberries. Inserting these numbers into the 2x2 table:

Ate Raspberries	Case	Control	TOTAL
Yes	21 (a)	4 (b)	25
No	9 (c)	56 (d)	65
TOTAL	30	60	90

$$\begin{aligned} \text{odds of exposure (cases)} &= 21/9 = 2.3 \\ \text{odds of exposure (controls)} &= 4/56 = 0.07 \\ \text{odds ratio} &= \frac{\text{odds of exposure (cases)}}{\text{odds of exposure (controls)}} = \frac{2.3}{0.07} = 32.7 \\ &\text{or} \\ \text{odds ratio} &= \frac{ad}{bc} = \frac{(21)(56)}{(4)(9)} = 32.7 \end{aligned}$$

Interpretation: The odds of exposure to raspberries was over 30 times higher among cases than controls.