Thermostable *Clostridium perfringens* as Cause of Food Poisoning Outbreak

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ALTHOUGH food poisoning due to *Clostridium perfringens* was first reported by McClung in 1945 (*1*), there have been few well-documented reports of the association of *C. perfringens* with food poisoning in the United States. Recently Nelson and associates described an outbreak affecting college students under circumstances similar to those described here (*2*). Other outbreaks attributed to *C. perfringens* were reported in 1959 (*3*), and several scattered outbreaks since then have been summarized by Nelson (*2*).

A food poisoning outbreak which began February 24, 1966, affected 366 college students fed through a common kitchen in a new dormitory complex at the University of Wisconsin. Salmonellae, shigellae, and staphylococci, usually incriminated in bacterial food poisoning, were not encountered, and heat-resistant *C. perfringens* was demonstrated as the cause and contaminated gravy as the vehicle. A preliminary report of this outbreak appeared in the *Morbidity and Mortality Weekly Report* of March 26, 1966, page 103.

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The epidemiologic study and statistical analysis of the outbreak was carried out with the help of Dr. Josef Preizler, Wisconsin State Board of Health, Madison, Charles Boettcher, Wisconsin State Laboratory of Hyiene, and the sanitarians of the Madison City Health Department.

Epidemiologic Studies

The students developed an illness characterized by diarrhea, abdominal cramps, and little or no fever. Illness was confined to students eating at three of the six dining halls that serve 3,000 students through a common kitchen. The incriminated food was served at the evening meal on February 23, 1966. Three dining halls served a choice of roast beef with gravy or fish. Three others served a choice of hamburger or fish, and all other foods served were common to the six dining halls. A total of 2,954 students ate the meal.

Questionnaires concerning the food eaten, time of onset of illness, and clinical symptoms were distributed to students who ate in the three suspect dining halls. Questionnaires were returned by 366 ill students, 344 of whom included time of onset; 740 questionnaires were returned by well students. The clinical illness lasted less than 24 hours and was characterized mainly by diarrhea. Abdominal cramps were reported by about half of the students, while nausea, vomiting, and fever were rare. The distribu-

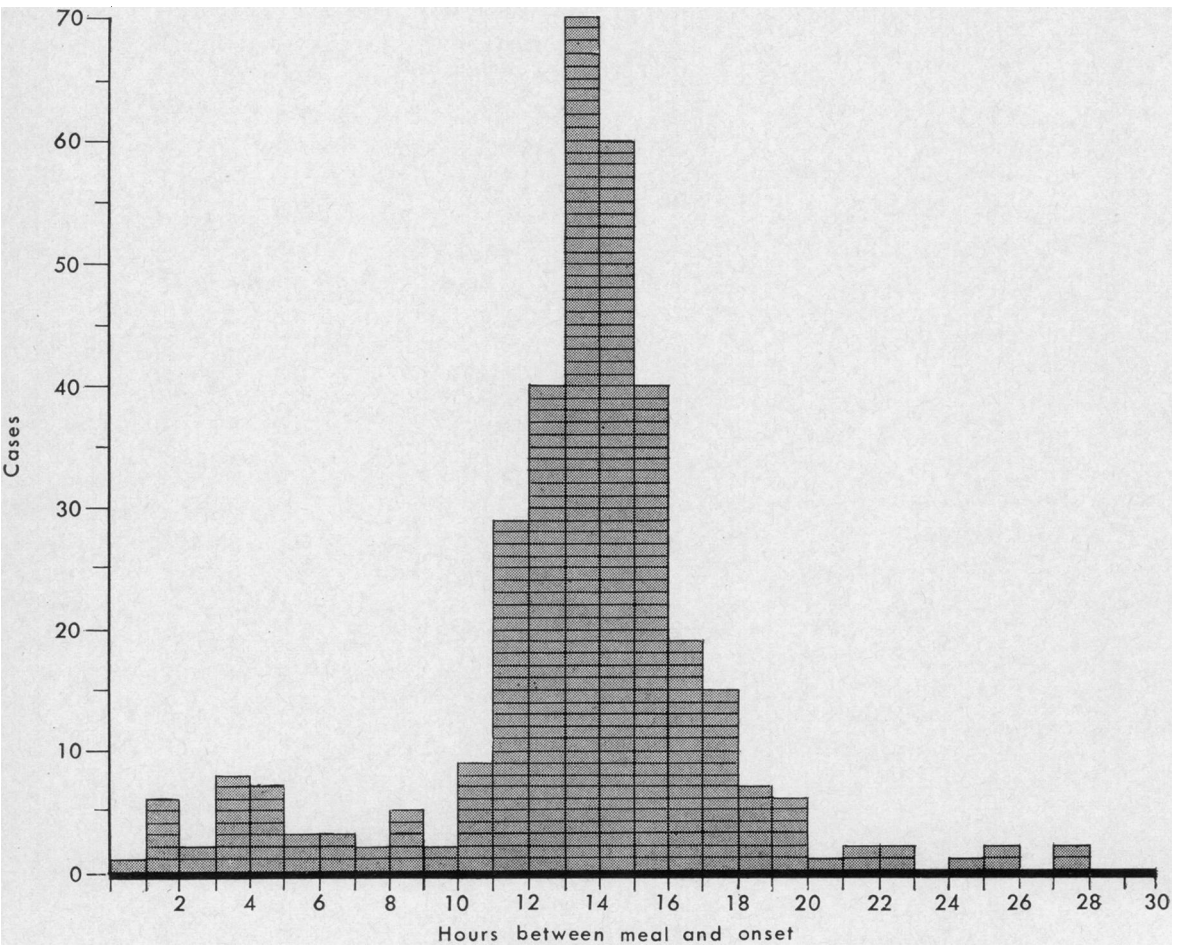
tion curve of the incubation period, shown in the chart, demonstrates a well-delineated peak at 14 hours with an abrupt rise and fall.

The attack rates among students who ate the different entrees at the evening meal are shown in the table. An attack rate of 69.9 percent among those who ate roast beef and gravy, 4.9 percent among those who did not eat roast beef and gravy, and the complete absence of illness in 48 students who ate roast beef without gravy incriminated the gravy as the source of the outbreak. According to the cooks who had prepared the evening meal, beef bone stock and gravy left over from February 22 had been added to fresh gravy made for the roast beef on February 23. Approximately 27 gallons of leftover gravy had been placed in three plastic containers, each holding about 9 gallons, and placed in the refrigerator overnight. The following day 7 gallons of freshly prepared gravy were added to the leftover material taken from the refrigerator; the entire mixture was brought to a rolling boil and served, separately from the roast beef.

Laboratory Studies

Although no leftover foods were available from the meal in question, samples of each food item, which are routinely collected and refrigerated for every meal, were available for culture. These were examined aerobically and anaerobically for the presence of enteric pathogens, staphylococci, and *C. perfringens*. None of these organisms were found in any of the food. The gravy was further examined for the presence of clostridial toxins by means of mouse-inoculation tests which proved negative. However, it is not known whether the tested gravy

Outbreak of food poisoning due to *Clostridium perfringens*, University of Wisconsin



sample included the leftover gravy or represented only the freshly prepared gravy.

Stool specimens were obtained from 20 ill students who had eaten the gravy and from 24 of the kitchen personnel, only 1 of whom admitted to illness. The stools were examined for *Salmonella* and *Shigella* using currently accepted procedures and for the presence of heat-resistant *C. perfringens* in the following manner. Stool specimens were inoculated into tubes of fluid thioglycollate broth (BBL), heated 1 hour in a boiling water bath, cooled under running tapwater, and incubated at 37°C. until growth occurred or up to 5 days. Tubes showing growth as indicated by gas production and the presence of large gram-positive bacilli were subcultured to blood agar plates and egg yolk agar plates (McClung and Toabe) and incubated anaerobically in a brewer jar at 37°C. for 24 hours. Colonies resembling *C. perfringens* were confirmed by the following characteristics: stormy fermentation in litmus milk; positive lecithinase reaction, which was neutralized by *C. perfringens* type A antitoxin; nonmotile; lack of visible spores; liquifaction of gelatin; production of H_2S; and acid and gas in glucose, sucrose, maltose, lactose, and glycerol but not in mannitol.

Stool specimens submitted by 19 of 20 ill students were positive for heat-resistant *C. perfringens* while the specimen from only 1 of 24 foodhandlers was positive. The foodhandler submitting the positive stool specimen was the foodtaster who had been ill during but not prior to the outbreak. Thirteen stools collected at the same time from healthy students who had not eaten the incriminated foods were examined for *C. perfringens* and all were negative. Five weeks after the outbreak a second stool specimen was obtained from 13 of the 19 students whose first specimen was positive for heat-resistant *C. perfringens*: only one was still positive. All 21 cultures were confirmed as *C. perfringens* type A by the Laboratory Branch of the Public Health Service's Communicable Disease Center in Atlanta, Ga., and all 21 cultures were found to agglutinate with Hobbs' type 10 antiserum.

Discussion

The clinical symptoms and epidemiologic aspects of the various *C. perfringens* outbreaks are quite characteristic and similar to those in this outbreak. The incubation period is usually 8 to 22 hours with a peak from 10 to 14 hours, and the illness is characterized by acute abdominal pain and diarrhea. Nausea and vomiting are rare. Fever, headache, and other signs of infection are seldom present. The illness is of short duration, usually 1 day or less.

The reported outbreaks are consistently associated with meat or some meat product such as gravy which may have been cooked some hours before eating and which, during the intervening hours, is stored at temperatures which would allow growth of the organism. The incriminated food usually will show large numbers of *C. perfringens* (often there are millions of organisms per gram of food), and it is sometimes possible to isolate the same Hobbs' serologic type from feces of the sick persons and from the food that they have eaten.

In the outbreak reported here, clinical symptoms and epidemiologic characteristics indicated that *C. perfringens* might be the etiological agent. Unfortunately no leftover food from the incriminated meal was available for bacteriological examination. Test samples of food which are routinely collected and refrigerated

Attack rate in University of Wisconsin students, *Clostridium perfringens* outbreak

Food	Ate food			Did not eat food		
	Number	Number ill	Percent ill	Number	Number ill	Percent ill
Fish	391	16	4.1	715	340	47.6
Hamburger	188	15	8.0	918	351	38.2
Beef with gravy	479	335	69.9	627	31	4.9
Beef without gravy	48	0	0	1,058	366	34.6

for each meal, however, were available for study and were negative for *C. perfringens*. However, samples of the leftover gravy incorporated in the new mix may not have been included in this routine sampling. Based on this experience, future samplings should include all food samples to be served, preferably from the serving line itself.

The isolation of *C. perfringens* from the stools of affected persons and not from controls in this study emphasized the importance of stool examinations. This is especially true when no food samples are available. Studies of both stool and food specimens should be a routine part of an investigation whenever possible.

The search for organisms like *C. perfringens* deserves emphasis because a large number of food poisoning outbreaks are reported annually in the United States in which no causative agent is identified. From 1957 to 1961, 50 percent of the food poisoning outbreaks in California were listed as "etiology unknown" (*4*). In 1959 there were 75 outbreaks of foodborne disease in which poultry and other meat were thought to be the vehicle of infection, but in which no etiological agent was identified (*3*). If one considers that 291 of 428 outbreaks in England associated with meat during the period 1959–63 were attributed to *C. perfringens* (*5*), it seems plausible to assume that a number of the 75 U.S. outbreaks of unknown etiology could have been caused by *C. perfringens*. It has been suggested that the dearth of reports of *C. perfringens* food poisoning in the United States may be the result of failure to include adequate anaerobic culture methods in laboratory investigations (*3*) and of the somewhat complex procedure required for the identification of food poisoning strains of *C. perfringens* (*6*).

However, the British have reported numerous outbreaks of *C. perfringens* (*welchii*) food poisoning. Hobbs (*7*) noted that over the 10-year period 1950–60 the number of reported outbreaks had risen to between 80 and 90 per year. It has been estimated that of the three main food poisoning organisms in England and Wales (salmonellae, staphylococci, and *C. perfringens*), *C. perfringens* has caused approximately one-quarter of all the food poisoning cases (*5*). The majority of the British outbreaks have been caused by nonhemolytic, heat-resistant strains of *C. perfringens* such as found in this study (*8*). In the United States, outbreaks have more often been attributed to heat-sensitive strains of the organisms (*9*) and too little attention may have been directed to the detection of heat-resistant strains in studies of these outbreaks. In the college outbreak recently reported by Nelson and associates (*2*), *C. perfringens* was cultured from lamb stew, but its heat resistance was not determined and fecal specimens from ill students were not cultured for *C. perfringens*.

Public health and other laboratories charged with identification of the etiological agents of food poisoning outbreaks are encouraged to develop appropriate routine anaerobic isolation and identification techniques if they are not already in use or to forward materials for study to laboratories which are set up for such studies. In this way a better understanding of the etiology of such outbreaks, as well as the relative role of *C. perfringens* and other anaerobes in their causation, may be achieved.

Summary

A large outbreak of acute gastroenteritis affected 366 students at the University of Wisconsin, Madison, who ate in a common mess. The main clinical features were diarrhea and abdominal cramps.

The mean incubation was 14 hours and there was a sharp rise and fall of the epidemic curve. Analysis of food questionnaires incriminated gravy as the source of infection with an attack rate of 69.9 percent among those who ate it. *Clostridium perfringens* was suspected as the cause, and this was confirmed by laboratory studies.

Heat-resistant *C. perfringens*, type A, Hobbs' type 10, was isolated from the stools of 19 of 20 ill students and from none of the 13 stools from healthy students eating in the same mess hall who did not eat the gravy; 1 of the 24 stool specimens from kitchen personnel was positive for the organism. The food samples were negative.

The use of appropriate anaerobic methods of culturing stool specimens and food samples should be a routine part of the bacteriological investigation of a foodborne outbreak of gastroenteritis. This practice would probably lead to increased recognition of outbreaks of food

poisoning due to *C. perfringens* and give a truer picture of the organism's relative importance among the causes of foodborne disease in the United States.

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Diplomate Recognition of Sanitarians

The American Intersociety Academy for Certification of Sanitarians, Incorporated, was established in March 1966 by the Sanitarians Joint Council, an organization composed of representatives of the National Association of Sanitarians, the International Association of Milk, Food and Environmental Sanitarians, and the American Public Health Association.

The new academy has three major objectives:

1. To improve the practice, elevate the standards, and advance the professional functions and ethical standards of practice of the professional sanitarian in environmental health.

2. To grant and issue to qualified sanitarians certificates indicating special knowledge, competence, and proficiency in various fields of environmental health. The fields in which certificates may be granted by the corporation are environmental sanitation (general), milk and food sanitation, vector and solid waste control, radiological health, air pollution control, industrial hygiene, institutional sanitation, water supply and waste disposal, housing hygiene, environmental health administration, and such other defined comprehensive fields as may be determined by two-thirds vote of the board of directors.

3. To provide for and conduct examinations to determine the fitness of applicants to become diplomates of the academy.

Officers of the board of directors are H. S. Adams, chairman, Indianapolis, Ind.; Dr. A. Harry Bliss, vice chairman, Berkeley, Calif.; Darold W. Taylor, secretary, Alexandria, Va.; and B. Russell Franklin, treasurer, Philadelphia, Pa. Other board members are Emill T. Chanlett, Chapel Hill, N.C.; E. R. Diddams, Springfield, Ill.; Larry J. Gordon, Albuquerque, N. Mex.; William C. Miller, Jr., Bethesda, Md.; A. Faegin Parrish, Decatur, Ga.; Verne C. Reierson, Portland, Oreg.; and Edwin L. Ruppert, Dallas, Tex.

Persons wishing more information about the academy should write to Darold W. Taylor, secretary, 2101 Wakefield Street, Alexandria, Va. 22308.

Public Health Service Staff Appointments

Dr. Paul Kotin, while continuing as scientific director for etiology in the National Cancer Institute, has been named director of the new Division of Environmental Health Sciences at the National Institutes of Health.

The Division conducts research on the effect of environmental hazards, provides grants for research and research training to scientists in colleges and universities, and contracts with universities, research institutes, national laboratories, and private industry for services needed to expedite work in the laboratories.

Dr. Kotin joined the National Institutes of Health in 1962, following more than 10 years of teaching at the University of Southern California School of Medicine where he was the Paul Peirce professor of pathology.

Dr. Kotin received his M.D. degree from the University of Illinois in 1940. He is a diplomate of the American Board of Pathology, a member of several committees and advisory groups concerned with environmental health problems, and a recipient of the Department of Health, Education, and Welfare Superior Service Award.

Dr. Gerald D. LaVeck has been appointed director of the National Institute of Child Health and Human Development. He succeeds Dr. Donald Harting who has joined the American Public Health Association as director of its family planning project. Prior to this appointment, Dr. LaVeck was acting scientific director of the Institute and for the past 3 years, director of the Institute's Mental Retardation Program.

Since coming to the National Institutes of Health in 1963, Dr. LaVeck has helped to develop the mental retardation activities of NICHD and NIH. He is also chairman of the NIH Staff Group on Mental Retardation. Recently he received a Superior Service Award from the Department of Health, Education, and Welfare for his efforts in the development of a broad and effective program for the conduct and support of research and training in mental retardation.

From 1957–58, Dr. LaVeck was an instructor in pediatrics and a fellow in mental retardation, University of Washington School of Medicine, Department of Pediatrics, Neurology, and Neuropathology. He was clinical director of the Rainier School, Buckley, Wash., 1958–62 and head of the Crippled Children's Service, Washington State Department of Health, 1962–63, and a clinical assistant and then associate professor of pediatrics at the University of Washington School of Medicine, 1960–63. Currently, Dr. LaVeck is a clinical associate professor of pediatrics at Georgetown University School of Medicine, Washington, D.C.

Born in Seattle, Wash., Dr. LaVeck took his B.S. degree in 1948 from the University of Washington and his M.D. degree in 1951 from the University's School of Medicine.

Dr. LaVeck is a diplomate of the American Board of Pediatrics, a fellow of the American Academy of Pediatrics and the American Public Health Association, and a member of the American Medical Association and the Washington State Medical Association.

Dr. Leonard D. Fenninger heads the new Bureau of Health Manpower in the Public Health Service. The Bureau is charged with stimulating the development of health manpower resources throughout the nation so that needed health services are available to all of the American people.

Before coming to the Bureau, Dr. Fenninger was director of the University of Rochester's Strong Memorial Hospital, holding this post since 1961. Previously he was associate dean of the University of Rochester School of Medicine and Dentistry since 1958.

Dr. Fenninger's principal research interests have been the study of health care patterns in the community, metabolism in cancer, and electrolyte metabolism. He is a member of a number of scientific and professional organizations, including the American Association for Cancer Research, the New York Academy of Sciences, the Rochester Academy of Science, and the Monroe County Medical Society.

Born in Hampton, Va., Dr. Fenninger attended secondary schools in Princeton, N.J. He received his bachelor's degree from Princeton University in 1938 and his M.D. degree from the University of Rochester in 1943.