Outbreak of Milk Poisoning Due to a Toxin-Producing Staphylococcus Found in the Udders of Two Cows*

JAMES A. CRABTREE, M.D.,
AND
WILLIAM LITTERER, M.D., PH.G., F.A.P.H.A.
Director, Division of Preventable Diseases, and Director, Division of Laboratories, Tennessee State Department of Public Health, Nashville, Tenn.

So far as the authors are aware, Barber (1914) first suggested that a toxin-producing staphylococcus was responsible for cases of food poisoning, when he described cases of gastro-enteritis which had occurred from time to time (1909–1913) on a certain farm in Nueva Ecija Province, Luzon, P. I., and attributable to the ingestion of improperly refrigerated milk which contained staphylococci in abundant quantities.

During the past three or four years outbreaks of food poisoning due to staphylococcus "enterotoxin" have been reported by a number of investigators, notably Jordon, and indeed the splendid work of Jordon and his associates has stimulated a widespread interest in the problem.

This report refers to a series of outbreaks of acute food poisoning occurring at Pleasant Hill Academy, Pleasant Hill, Tenn., from time to time during the period July 27–October 20, 1933.

Pleasant Hill Academy is a preparatory school operated under the auspices of the American Missionary Association, and situated on the Cumberland Plateau in Cumberland County, Tenn., 90 miles west of Knoxville and 120 miles east of Nashville.

The school owns a farm of several hundred acres. A herd of 13 cows supplies all of the milk. Practically all of the labor at the school is done by the students. They work on the farm, cook, wait on table, fire furnaces, do janitor work, and operate the laundry.

The principal and his family reside in a separate house on the school campus, and the farm-manager occupies an apartment in one of the dormitories, and for these two families meals are prepared and served within their respective homes. The remainder of the faculty members and all of the resident students live in the dormitories and eat in one dining room, the arrangements being such that one faculty member is placed with each table of students. Thus, with the exception of the principal and farm-manager, the faculty and student body have the same food supply.

During the period June–August, 1933 (school vacation), the only persons who lived on the school premises were the

* Read before the Epidemiology Section of the American Public Health Association at the Sixty-third Annual Meeting in Pasadena, Calif., September 5, 1934.

families of the principal and the farm-manager and 2 students who helped with the operation of the farm and dairy, these students staying in the dormitory and boarding with the farm-manager.

On July 27, 1933, cases of acute poisoning occurred in all of the 6 members of the household of the farm manager, including the 2 students. Clinically, the cases were characterized by acute and sudden onset of nausea and vomiting, and in some instances prostration. Purging and diarrhea did not occur in these cases, but were prominent symptoms in later outbreaks. All cases were reported to be afebrile, and recovery rapidly followed emptying of the stomach. All of the cases had onset within 15 minutes of each other, the time being about 10 a.m., or approximately 3 hours after breakfast.

School opened during the last week of August, 1933, when 88 resident students enrolled, and these, with 9 faculty members, made a total resident population of 97, ranging in age from 14 to 54 years, and including 51 females and 46 males.

On September 4, 1933, the second outbreak occurred involving 45 cases, all having onset at about 9 p.m., or approximately 3 hours after the evening meal. In this and in all subsequent outbreaks no cases occurred in the family of either the principal or the farm-manager. Subsequent outbreaks occurred on September 13, 20, 27, October 2, 10, and 20.

It is interesting that the 7 outbreaks which occurred following the opening of school were rather evenly distributed with respect to time, the intervals between successive outbreaks ranging from 7 to 10 days.

A total of 242 cases occurred, 233 in students and 9 in faculty members. Students suffered an attack rate 2.9 times that of faculty members, the rate for the former being 2.9 cases per person and for the latter 1 case per person. In 2 of the outbreaks the approximate hour of onset of cases was 9 p.m. and for the remaining 5 was 10 a.m.

Table I shows for each of the 7 outbreaks the date, the approximate hour of onset of the cases, and the number of

TABLE I

DATES OF OUTBREAKS OF FOOD POISONING, APPROXIMATE HOURS OF ONSET OF CASES, AND NUMBER OF CASES WITH CASE RATES IN STUDENTS AND FACULTY MEMBERS AT PLEASANT HILL ACADEMY, TENN.—SEPTEMBER 4–OCTOBER 20, 1933

Date of Outbreak	Approximate Hour of Onset	Number of Cases			Case Rate (=100)		
		Students	Faculty	Total	Students	Faculty	Total
Sept. 4	9 P.M.	44	1	45	50.0	11.1	46.4
" 13	10 A.M.	30	2	32	34.0	22.2	33.0
" 20	10 A.M.	15	0	15	17.0	—	15.5
" 27	10 A.M.	25	1	26	28.4	11.1	26.8
Oct. 2	9 P.M.	34	2	36	38.6	22.2	37.0
" 10	10 A.M.	16	0	16	18.2	—	16.5
" 20	10 A.M.	69	3	72	78.5	33.3	74.5
Total		233	9	242	290.0	100.0	250.0

cases with case rates in students and in faculty members.

The explosiveness of the outbreaks, with all cases having onset within 15 to 30 minutes of each other, indicated that the etiologic agent was transmitted from a common source; and the consistency with which onset of cases occurred 3 hours following a meal further indicated that food was the most probable vehicle.

The absence of temperature, the abrupt and violent onset, the short duration, and rapid recovery without material after-effects all tended to indicate that the illness was due to a toxemia or poisoning rather than an infection.

The cases were not reported to the State Health Department until October 3, on the morning following the 5th outbreak. Investigation was made by one of us (J. A. C.) at that time, when epidemiological case records were obtained for all of the cases which had occurred previously. Subsequent cases were investigated within less than 24 hours following onset.

In the students the distribution of cases with respect to sex and age was in no way unusual. In the faculty members, the numbers were too small to justify comparisons.

Since a total of 242 cases occurred in 97 persons, it would follow that a number of persons suffered repeated attacks. The distribution of persons according to the number of attacks is shown in Table II.

TABLE II

DISTRIBUTION OF ATTACKS

Number of Attacks	*Number of Persons*	*Per cent of Total*
5	12	12.4
4	16	16.5
3	19	19.6
2	26	26.8
1	9	9.3
0	15	15.4
	97	100.0

Because of the practice of the school of assigning one faculty member for each group of students at a table, both students and faculty members ate the same food, which was served and prepared by the same people.

Information was obtained as to the various articles of food consumed by each of the 97 persons at the meals immediately preceding onset of the cases.

With the exception of milk, no article of food eaten during these meals appeared more frequently in the diet of those ill than of those not ill.

Table III shows for each of the outbreaks the number of cases, and case rate in persons who drank milk during the meal immediately preceding onset of cases, and for comparison the attack rate in those who did not drink milk.

Out of the 242 cases, all but 1 occurred in persons who had drunk milk during the meal immediately preceding onset. The attack rates in milk consumers varied from 17.6 per cent on September 20, to 81 per cent on October 20. In view of subsequent data which clearly established the fact that milk was the vehicle of the causative agent, it is quite likely that for the case occurring in the person who denied drinking milk, either the history was unreliable, or perhaps the illness may have been of another nature.

Following the outbreak of October 2, fecal specimens from all of the 37 persons who were in any way connected with the handling of food were examined for the typhoid, dysentery, and salmonella group of organisms with entirely negative results.

Following the outbreak of October 20 all articles of food served at the meal immediately preceding onset, and also pooled specimens of first vomitus of the patients were examined both bacteriologically and chemically. In addition, specimens of throat secretions from all of the food handlers were immediately streaked on rabbit blood-agar plates and

TABLE III

CASES OF FOOD POISONING AND ATTACK RATES IN PERSONS WHO:
(A) DRANK MILK AT THE MEAL PRECEDING ONSET
(B) DID NOT DRINK MILK AT THE MEAL PRECEDING ONSET

Date of Outbreak	Drank Milk at Meal Preceding Onset			Did Not Drink Milk at Meal Preceding Onset		
	No. Persons	Cases		No. Persons	Cases	
		Number	Attack Rate (=100)		Number	Attack Rate (=100)
Sept. 4	85	45	53.0	12	0	0.0
" 13	89	32	36.0	8	0	0.0
" 20	85	15	17.6	12	0	0.0
" 27	88	25	28.4	9	1	11.1
Oct. 2	83	36	43.4	14	0	0.0
" 10	88	16	18.2	9	0	0.0
" 20	89	72	81.0	8	0	0.0

brought into the laboratory for bacteriological examination. The articles of food included cereal, sugar, toast, coffee, milk, bacon, scrambled eggs, apple sauce, and butter. The chemical examination * included a search for arsenic, lead, strychnine, mercuric chloride, barium and antimony salts, phosphorus, and various alkaloids. Results of chemical analyses were entirely negative.

In the bacteriological examination, each article of food and the vomitus were examined directly by stained smear (Gram) and a loopful was planted for culture on plates of plain agar, Endo's, eosin-methylene blue agar, and 3 per cent rabbit blood-agar.

All of the articles of food except milk contained only very few bacteria, and these were of various types.

From the milk and the vomitus, hemolytic staphylococci were obtained in enormous numbers and practically in pure culture. The staphylococci were decidedly hemolytic and were of both the albus and aureus types, the ratio of aureus to albus being about 40 to 1.

Of the 37 food handlers whose throat secretions were planted on rabbit blood-agar plates, 12 showed large numbers of hemolytic staphylococci, both aureus and albus, the aureus predominating in a ratio of 50 to 1. Incidentally, all of these 12 persons had suffered attacks of poisoning less than 24 hours before the specimens were obtained. These 12 persons were reëxamined at 3 day intervals during the following 3 weeks, and in all of them the organisms had disappeared apparently by the end of this time. Each succeeding examination showed a diminished number of organisms, and in all but 3 persons, results of examinations were negative at the end of 12 days.

The staphylococci recovered from the milk, the vomitus, and the throat secretions of the food handlers were tested for production of toxic products, the

* Chemical examinations were made by Dr. J. W. Sample, Chief Chemist, Tennessee State Department of Agriculture, Nashville.

technic employed being that described by Woolpert and Dack (1933).

In testing for the presence of enterotoxic substance in the filtrates from these organisms only human volunteers were employed. Three c.c. of filtrate in approximately 3 ounces of pasteurized milk were given to persons who had not eaten any food for more than 5 hours. Where illness followed the ingestion of these filtrates, the incubation period was shorter (though somewhat variable for different filtrates), and symptoms were less severe, but otherwise clinically identical with those occurring in the outbreaks.

Although a sufficient number of human volunteers could not be obtained to test all of the filtrates that were prepared, some of the organisms recovered from the milk, the vomitus, and the throat secretions of the food handlers were tested.

The yellow staphylococci recovered from the milk, the vomitus, and from 5 of the 12 food handlers proved to be toxin-producers, the filtrates producing illness in all of the human volunteers. Filtrates from the organisms isolated from the milk and the vomitus produced symptoms within 30 minutes, but those from the 5 food handlers produced illness with incubation periods ranging from 1½ to 3 hours.

In these tests, 8 persons were used as controls, these being fed milk and sterile media, and none of them became ill.

All epidemiological evidence, supported by bacteriological findings, indicated that milk was the most probable vehicle of the "enterotoxin." However, it was necessary to determine the source of the organisms which apparently produced the "enterotoxin"; *i.e.*, whether the secreted milk itself contained the organisms, or contamination occurred through handling of the milk by one or more infected food handlers. The latter possibility was suggested by the fact that 12 of the food handlers were found to harbor the organisms in their throats.

Beginning October 23, specimens of milk were obtained under as nearly aseptic conditions as possible from each of the 13 cows, at a rate of 3 specimens per week (every other day) for a period of 5 weeks. During this period, specimens from 2 cows (Nos. 2 and 7) showed almost consistently a large number of hemolytic staphylococci. Specimens from 4 other cows occasionally showed the organisms, but filtrates from these when fed to human volunteers failed to demonstrate enterotoxic substances in 50 c.c. amounts.

Of the 12 specmens from cow No. 2, 9 contained hemolytic staphylococci, and from Cow No. 7, 12 of the 15 specimens were positive. Both the aureus and albus types were found in the milk from each cow, the aureus predominating in proportions, ranging from 25 to 1, to 60 to 1. Filtrates from the albus produced no symptoms when fed in 50 c.c. amounts to human volunteers. Filtrates from 7 different aureus cultures produced illness in 12 of 15 human volunteers when fed in 3 c.c. amounts.

The number of organisms to the c.c. of milk from cow No. 2 averaged about 2,900, and from cow No. 7, about 4,500. One specimen showed a count as high as 17,500, while others contained only a few. A negative specimen would usually be followed by a tremendous increase to the maximum bacteria count. Table IV shows the results of examinations of the various specimens of milk taken from cows Nos. 2 and 7 over the 5 week period.

Evidently somewhere in the udders of these 2 cows there were pus pockets which would occasionally break loose, producing a "showering" of staphylococci, followed by a gradual diminution in number of organisms for a time, after which another "showering" would take place. This recurrent "showering" of organisms is of particular in-

TABLE IV

RESULTS OF EXAMINATIONS OF SPECIMENS OF MILK TAKEN FROM 2 COWS AT PLEASANT HILL ACADEMY, TENN., OVER THE PERIOD OCTOBER 23 TO NOVEMBER 27, 1933

Date	Number of Hemolytic Staphylococci per Cubic Centimeter of Milk	
	Cow Number 2	Cow Number 7
Oct. 23	2,500	1,500
" 25	1,500	200
" 27	100	0
" 31	50	0
Nov. 3	5	17,500
" 5	0	14,200
" 8	0	5,000
" 10	14,000	1,000
" 13	8,000	75
" 15	5,000	0
" 18	300	15,700
" 20	0	9,600
" 23	No specimen	1,800
" 25	No specimen	150
" 27	No specimen	25

terest in view of the recurrent and more or less evenly spaced outbreaks of poisoning.

These 2 cows were examined by a veterinarian who stated that they had " Garget " or mastitis which is a chronic infectious disease characterized by recurrent acute attacks when the cow is drying off, when she freshens, when she is fed heavily on a high protein diet, or when she is otherwise exposed to unusual strain. If the disease is acutely active, the milk may be changed in appearance. The milk from both of these cows was at times ropy and stringy, containing red blood cells and numerous pus cells. From cow No. 7, the milk on two occasions showed the presence of macroscopic blood.

The school dairy consisted of 13 cows, tuberculin tested annually and apparently in good physical condition. The dairy barn is of reasonably modern construction, with individual stalls for the cows, cement floor, and ample water supply.

The milking is done by hand, by 5 boys who are students at the school. Generally speaking, sanitary precautions concerning the milkers, the cows, and the milk utensils could be regarded as satisfactory.

Each milker used an ordinary 1 gallon milk pail from which the milk was transferred, at the dairy barn, directly into 5 or 10 gallon cream cans. In this transfer the milk was strained through clean cheesecloth.

With the exception of that which went to the residences of the school principal and farm-manager, all the milk was carried from the barn to the kitchen (in the girl's dormitory) in these large cream cans, and stored in these containers until it was served. The kitchen contained two refrigerators, one an ordinary commercial Frigidaire and the other a built-in cold storage room, both being refrigerated by means of one electrical motor located in the basement of the kitchen. The cold-storage room was used largely for the storage of meat and these large containers of milk. The temperature in this room was considerably higher than that in the commercial Frigidaire.

The milk which was brought in at night was stored over night and served the following morning. The morning's milk was usually served at the evening meal.

The milk which went to the residences of the school principal and farm-manager was carried directly from the dairy barn to the residences in the small milk pails, transferred to milk con-

tainers and immediately placed in the refrigerators in the respective residences. Because of the much smaller milk containers and the lower temperatures in the refrigerators, the milk for the families of the school principal and farm manager became chilled very much more quickly and was brought to a lower temperature than that stored in the large cream cans and served to the rest of the resident population. This rapid chilling presumably inhibited the growth and toxin-production of the staphylococci present in the secreted milk, and therefore offers the most plausible explanation of the fact that no case of poisoning occurred in any member of these two households, notwithstanding the use of milk from the same cows that supplied the rest of the faculty and student body.

Recommendations to the school authorities embodied removal of 2 of the cows from the herd, and measures directed toward improvement of refrigeration facilities. These recommendations were put into effect immediately, and no additional cases of poisoning have been reported to date.

DISCUSSION

We believe that this is the first recorded outbreak of poisoning due to enterotoxic products of the staphylococcus where it was possible both to identify the enterotoxic substance, and to determine the source of the staphylococcus.

The circumstances under which the outbreak occurred merely add to already overwhelming evidence supporting the need for rigid sanitary control of milk supplies.

REFERENCES

Barber, M. A. Milk Poisoning Due to a Type of Staphylococcus Albus Occurring in the Udder of a Cow. *Phillipine J. Sc.*, 9, 6:515 (Nov.), 1914.

Woolpert, O. C., and Dack, G. M. The Relation of Gastro-Intestinal Poison to Other Toxic Substances Produced by Staphylococci. *J. Infect. Dis.*, 52, 1:6–19 (Jan.-Feb.), 1933.

Maternity Benefits in Cuba

A DECREE signed by the President of Cuba on April 18, 1934, prohibits the employment of women for 6 weeks following confinement. The prohibition applies to all establishments, whether industrial or commercial, public or private.

According to this decree a pregnant women is permitted to leave her employment 6 weeks before the probable date of childbirth, upon presentation of a physician's certificate. For the duration of her absence from work the woman is to receive a cash benefit equal to her wages, paid from an insurance fund to which the State, the employers, and the women workers will be required to contribute. Attendance by a physician or midwife will also be provided.

An employer must permit every nursing mother to have two daily periods of half an hour each in which to nurse her child, exclusive of the rest periods provided by law.

Pregnancy or illness connected with it must not constitute a cause of discharge, unless the woman's absence exceeds the time allowed for that purpose.

No pregnant woman may be employed on strenuous work or on work that may interfere with the normal development of the child.

Every industrial or commercial establishment employing 50 or more women must provide a day nursery for employees' children under the age of 2.

Fines are provided for violations.—*Gaceta Oficial de la Republica de Cuba*, April 20, 1934.