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A Long-term Carrier of Cholera: Cholera Dolores

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The first known long-term carrier of cholera, found in the Philippines, is described. The carrier, Dolores M., who had suffered from El Tor cholera in August 1962, continued intermittently to excrete vibrios of the same characteristics as the original isolates until the date of reporting (1966). Duodenal intubation proved that the vibrios are lodged in her biliary tract. Her serum antibody titre continued to remain high in the absence of vaccination against cholera.

It is common knowledge that cholera patients or contacts usually get rid of the invading *Vibrio cholerae* within a very short time of their encounter with it (Pollitzer, 1959). A chronic carrier state for cholera comparable to that for typhoid fever has never been proved.

A few cases of cholera El Tor in the Philippines found to excrete vibrios intermittently 3 to 4 months after cure (Dizon, 1962) were followed closely. Most of them became negative for agglutinable vibrios on repeated examination. Five of them were negative on duodenal intubation and had low serum antibody titres in 1965. Dolores M., usually referred to as Cholera Dolores, has, however, proved to be the solitary exception.

Dolores M. is a 46-year-old married female with 6 children. She is a housewife in a typical Philippine family of modest income. During the epidemic of 1962, when she had been carrying her sixth child for 5 months, she suffered from a mild attack of cholera El Tor, was admitted to the Provincial Hospital of Negros Occidental on 28 August 1962 and was discharged on 7 September 1962, when her stool report was still positive for El Tor vibrios. She was not treated with antibiotics.

She allegedly suffered from jaundice soon after the attack of cholera. For about 3 months beginning May 1965, she complained of occasional epigastric pains.

EPIDEMIOLOGICAL HISTORY OF THE CASE

Deogracias M., husband of the patient, had diarrhoea on 21 August 1962, but he was not ill enough to be hospitalized and investigated. At this time, cholera resurgence in the area was at its peak, and he often brought seafood from Bacolod, which was eaten uncooked by the whole family.

Three days after his father, Rodrigo, a son 6 years of age, had diarrhoea and vomiting; the next day, Rogelio, another son, 4 years old, had diarrhoea but did not vomit.

Two days after Rogelio, Dolores M. (the mother) became ill and was confined at Negros Occidental Provincial Hospital the next morning. Another boy, aged 2, had diarrhoea but no vomiting.

Except for the mother, no sick members of the family needed hospitalization, so no rectal swabs were taken at that time. However, Rodrigo was found positive on 28 August when the household contacts of Dolores M. were swabbed.

CLINICAL STATUS OF THE CARRIER IN 1966

She had no complaints. She looked pale on general physical examination but no other significant abnormality could be detected.

Her blood, examined on 10 February 1966, revealed Hb, 60%; RBC, 2.3 million/mm^3; WBC, 7500/mm^3 with 5% eosinophils.

Her liver-function test on 25 November 1965 showed: total bilirubin, 0.9 mg% (direct 0.5 mg%, indirect 0.4 mg%); thymol turbidity test, 4.25; SGPT, 30 units/ml; CCFT, negative at 24 hours.

Gastric juice analysis on 25 November 1965 revealed free HCl, 9°; total acidity, 19°.

Oral cholecystography on 25 November 1965 showed no stone or shadow of gall bladder. The radiologist diagnosed chronic cholecystitis.

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BACTERIOLOGICAL FINDINGS ON STOOL OR RECTAL SWAB EXAMINATION

Dolores M. was subjected to repeated examination of stools and rectal swabs at frequent but irregular intervals; the results are shown in Table 1. Swabs collected while she was in the hospital are also included. All these isolates were similar in their characteristics. They were serologically Ogawa in type, and they fermented sucrose and mannose, but not arabinose. The strains caused haemagglutination of chicken red cells; were haemolytic to sheep cells when grown in nutrient broth, heart infusion broth (HIB) and heart infusion broth with 1% glycerol; were resistant to 50 units of polymixin B (Difco) and also to cholera group-IV phage at routine test dilution. It may be noted that this chronic carrier was excreting haemolytic agglutinable vibrios when almost all the isolates from the cholera patients and contact carriers in the locality were non-haemolytic when grown in nutrient broth or HIB. None of her strains reacted with the rough serum. On two occasions in 1965, her stool was quantitatively examined for vibrios, when she was found to excrete about 2×10^3 and 2×10^4 vibrios per g of stool.

SEROLOGICAL FINDINGS

In October 1964, in February and November 1965, and in February 1966, the serum was examined for agglutinating antibody using live antigen (Barua & Sack, 1964); the titre was about 1: 640 to 1: 1280.

BACTERIOLOGICAL FINDINGS OF THE SPECIMENS OBTAINED BY DUODENAL INTUBATION

Sample A collected on 25 November 1965 (Table 2)

Pre-egg sample (pH 7.0–7.4). On viable count about 110 agglutinable vibrios per ml were found. All the colonies were typical and smooth when examined both by the naked eye and with a stereoscopic microscope.

Post-egg sample (pH 7.2). About 870 viable organisms per ml were detected. Of the 87 colonies produced on the plate inoculated with 0.1 ml of the fluid, 61 colonies were typical and smooth, but 26 were opaque. These latter colonies were subsequently found to be the rugose variants. They produced yellowish opaque colonies with a corrugated surface on the nutrient agar plates. The irregular and radial corrugations, opacity and pigmentation increased with age. The colonies were

TABLE 1
RESULTS OF BACTERIOLOGICAL EXAMINATION

Dates on which the vibrio could be isolated	Dates on which the vibrio could not be isolated
1962	
28–31 Aug.	
1–7, 29 Sept.	
13, 28 Oct.	7 Oct.
4, 22, 30 Nov.	
16, 30 Dec.	
1963	
13 Jan.	6, 28 April
23 March	5, 12, 19, 25 May
21 April	14, 22 Sept.
28 Sept.	13, 20, 26 Oct.
24 Nov.	3, 10 Nov.
1964	
21 July	9 June
3 Nov.	30 July
	4, 10, 18 Aug.
	10 Oct.
	10, 23 Nov.
	9, 24, 26 Dec.
1965	
2 Jan.	10 Jan.
22 Feb.	1, 15, 25 Feb.
11, 18, 19, 21, 28, 30 June	5, 18, 26 March
5, 19, 20 July	5, 11, 20, 29 April
	10, 12, 30 May
	30 June
	3, 19, 22 July
	9 Aug.
	14, 27 Oct.
	24 Nov.
1966	
18 Jan.	21, 22, 27 Jan.
	4, 5 Feb.

TABLE 2

QUANTATIVE EXAMINATION OF DUODENAL FLUID COLLECTED 25 NOVEMBER 1965 (A) AND 10 FEBRUARY 1966 (B)

Sample	No. of agglutinable vibrios/ml	Characteristics of the colonies
A. Pre-egg sample	110	Typical and smooth
Post-egg sample	870	30% colonies were rugose
B. Pre-egg sample	Direct culture negative. Positive after enrichment	Typical and smooth
Post-egg sample	1 700	Typical and smooth

tough and adherent to the agar and dispersed with difficulty in normal saline. The dispersed organism reacted with the type-specific Ogawa and group O-1 anti-cholera serum but not with the rough serum. On repeated subculture they threw out irregular numbers of smooth colonies. The rugose variants had the same characteristics as the original smooth isolates.

Kappa-type phage (Takeya & Shimodori, 1963) was detected in some of the isolates from this chronic carrier. It was also found in the bile obtained by duodenal intubation.

Sample B collected on 10 February 1966 (Table 2)

Pre-egg sample (pH 7.2). Hookworms were found in the fluid. Direct culture of the fluid for vibrios gave no growth, but after enrichment the culture was positive for El Tor vibrio; quantitation was thus not possible.

Post-egg sample. Many hookworms were seen. Quantitative culture revealed about 1700 viable El Tor vibrios per ml of the fluid. It is interesting to note that, unlike the growth after the first intubation, there was no rugose variant. The organisms were smooth and typical El Tor vibrios having the same characteristics as those of the original isolates.

PATHOGENICITY FOR INFANT RABBITS

Strains taken from a case of cholera, 2 contacts, and 2 different isolates from Dolores M., the chronic carrier, were injected in different doses into the upper part of the small intestines of 8-day-old suckling rabbits. The strains employed for the test were kept on agar slants at room temperature for not more than 2 months.

The strains from the cholera case and the 2 contact carriers produced typical changes in the intestines with or without diarrhoea in 3 out of 5 and 4 out of 4 rabbits, as expected after introduction of a virulent strain, when the animals were challenged with 10^3 to 10^4 colony-forming units (CFU) or viable organisms respectively. One isolate from the chronic carrier produced no effect even when 10^8 CFU were injected, while another isolate from her produced typical changes at the same dosage in 2 out of 3 animals.

DISCUSSION

The findings reported here indicate that the chronic carrier state has been proved in this individual though she was an intermittent excretor. She does not appear to be a case of reinfection as the characteristics of the isolates over a period of 42 months have been found to be constant. While the prevalent strains in the locality were non-haemolytic to sheep cells when grown in nutrient broth, the isolates from Dolores M. continued to be haemolytic. Her serum antibody titre was also persistently high in the absence of vaccination; this titre usually declines to a low level within 6 to 12 weeks after recovery (Barua & Sack, 1964).

The results of tests of the fluid obtained by duodenal intubation on 2 occasions clearly indicate that the organisms are harboured in the gall bladder or somewhere in the biliary tract, because the number of organisms in the duodenal fluid obtained after the fatty meal was much more than the sample obtained before the meal.

The presence of *V. cholerae* has repeatedly been demonstrated in the gall bladder and biliary tract of cholera victims by many investigators in the past, but it was difficult to prove that this was an *ante mortem* phenomenon (Pollitzer, 1959). The associated pathological changes in the gall bladder could be pre-existing lesions and need not be taken as evidence of *ante mortem* invasion by vibrios. The isolated instance of excretion of *V. cholerae* by a cholera patient whose stool became negative for vibrios, through the fistula after operation for cholecystitis (Valk, 1915; quoted by Pollitzer, 1959), indicates that vibrios can infect the gall bladder during life. However, excretion in this case was only for 4 weeks. The chronic carrier state for cholera as described in the present paper has not been recorded previously.

The strains isolated from the carrier are very sensitive to the low concentration of chlor-

amphenicol, tetracycline, and streptomycin, but the carrier has not been treated so that the natural course of the infection may be observed. It is interesting to note that she has chronic cholecystitis and the administration of iodine containing dye did not sterilize the gall bladder, as was indicated by the results of subsequent intubation and stool examinations.

Strains from carriers are generally believed to be rough in character, but the isolates taken from the present carrier were invariably smooth when tested with rough serum, although rugose variants were obtained from her bile after the first duodenal intubation. It is possible that rugose variants in her stools were missed, as such colonies were not specially looked for. These rugose variants were not found in the bile by the second intubation and may not be responsible for the chronic carrier state.

The virulence in infant rabbits of 2 recent isolates from this long-term carrier was compared with that of the isolates from a cholera case and 2 contact carriers by injecting these organisms into the upper part of the small intestine when the animals were 8 days old. As indicated, the 2 isolates from the long-term carrier varied in their capacity to cause disease in the infant rabbit, but these results may not reflect their effect on man.

Incidentally, it may be noted that the carrier was suffering from hookworm disease with anaemia, which may not have any relation to her carrier state except to lower her general resistance. Aside from chronic cholecystitis, no systemic abnormality was detected that could be responsible for her long-term carrier state.

Epidemiologically, it has not been possible to incriminate the carrier directly in any case of cholera. She lives in a relatively isolated house with her family in an unsanitary environment with a private water-source for domestic purposes.

Most of the family members have probably been exposed to cholera infection along with the carrier or subsequently and have become immune. Natural experiments like those that clearly pointed to the dangerous carrier status of " Typhoid Mary " did not take place in the rural Filipino environment of this carrier.

The limited examinations of her family members for vibrios have been negative. But so little is known of the factors that control the successful transmission of vibrios. She, with limited chance of transmission, appears to be harmless in the endemic area where she lives, but the impact of her presence in a suitable environment among a susceptible population with good chance of transmission might be entirely different.

RÉSUMÉ

En général, les vibrions cholériques disparaissent de l'organisme des malades ou des contacts peu de temps après l'avoir envahi. Jusqu'à l'observation du cas décrit dans cet article, on n'avait jamais mis en évidence, pour le choléra, d'état de porteur chronique comparable à celui qui existe pour la fièvre typhoïde.

Dolores M., couramment appelée Dolores Choléra, est porteuse de vibrions El Tor depuis 1962. C'est une mère de six enfants, âgée de 46 ans, appartenant à une famille philippine typique au revenu modeste. Pendant l'épidémie de 1962, elle a subi une atteinte bénigne de choléra El Tor, a été admise à l'hôpital le 28 août 1962 et en est sortie le 7 septembre 1962, alors que ses selles contenaient encore des vibrions El Tor. Elle n'a pas été traitée par les antibiotiques. A partir de cette date jusqu'au moment de l'enquête (1966), elle a continué à excréter de façon intermittente des vibrions ayant les mêmes caractéristiques que les premiers vibrions isolés. Il ne semble pas que ce soit un cas de réinfection, car les souches de vibrions isolées dans le voisinage ont varié pendant la même période. Un tubage duodénal a révélé que les vibrions s'étaient logés dans les voies biliaires. Le titre d'anticorps sériques est resté élevé en l'absence de toute vaccination anticholérique.

Son mari et deux de ses fils ont souffert de diarrhée avant qu'elle ne soit hospitalisée, et un autre de ses fils après son admission. L'un des deux fils a été trouvé porteur de vibrions cholériques lors des prélèvements effectués sur les contacts familiaux de Dolores M. le jour de son hospitalisation. Ni elle, ni les membres de sa famille n'ont présenté de symptômes de choléra depuis son retour de l'hôpital, et aucun membre de sa famille n'a été trouvé porteur de vibrions depuis cette date. On ne peut lui imputer directement la responsabilité d'aucun cas de choléra. Elle habite avec sa famille une maison relativement isolée et dispose de sa propre source d'eau pour les usages domestiques. Dans un milieu favorable et au sein d'une population réceptive, sa présence aurait pu avoir des conséquences totalement différentes.

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