

# Brief communications

## *Vibrio parahaemolyticus* serotypes in Calcutta, India

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### Abstract

A study of *V. parahaemolyticus* strains isolated from different sources in Calcutta showed that the serotypes associated with human gastroenteritis do not frequently coincide with those of marine and non-marine isolates. Some atypical, complex, and new serotypes are reported.

In Japan, the common serological types of *Vibrio parahaemolyticus* causing food poisoning are prevalent in sea fish and marine water (4). No parallel study in any other country has yet been described in the literature. *V. parahaemolyticus* seems to pose special problems in Calcutta because it occurs in fish and water of non-marine origin (1). In the present study, *V. parahaemolyticus* serotypes associated with sporadic diarrhoea were compared with those from potential marine and non-marine sources in Calcutta.

Since *V. parahaemolyticus* is facultatively halophilic, it is of interest to note that the salinity of samples of water from 10 different ponds in Calcutta was found to vary considerably (120, 220, 290, 440, 470, 550, 630, 650, 650, and 830 mg/litre as chloride ion). The ponds from which isolates were prepared for this study do not connect with sewerage, latrines, or river system, and are used for domestic purposes.

### Material and methods

**Strains.** The non-marine sources comprised 27 strains from small pond fishes and 25 strains from pond water. The marine reservoir was represented by 60 strains from water from the river Hooghly and 53 from sea shrimps. Sixty-two strains from human diarrhoea were also included. The procedure for the isolation and screening of strains from different sources has been described by Chatterjee (1).

**Serotyping.** K antigen type determination was

done by slide agglutination of 18–24-hour agar culture suspensions in 2% saline solution, using 52 individual K antisera following prior testing with 8 pooled antisera. The O groups were ascertained by agglutination tests on slides with 11 antisera after autoclaving each suspension for 0.5, 1, and 2 h. The set of Toshiba *V. parahaemolyticus* antisera employed in this study was kindly donated by Professor Takeshi Yokota, Juntendo University, Tokyo, Japan. A new K antiserum was prepared according to the methods of Edwards & Ewing (2).

### Results and discussion

Table 1 shows the serotypes identified from strains from different sources. The majority of the faecal isolates were serotypable but not so many of the marine and non-marine isolates could be typed. The serotype 05: Cal/Ka, a new K antigenic type, appears to occur in a significant proportion of diarrhoea samples in Calcutta. Of the types detected in human diarrhoea, 01: K56, 04: K55, 05: K30, and 06: K46 were not reported from Japan and the United States (4, 5). Sakazaki et al. (4) concluded that an individual K antigen is invariably linked with one particular O group. This postulate needs modification in view of the presence of atypical types in the Calcutta series. That a strain may reveal 2 K antigens (i.e., that it may be of a complex type) is intriguing.

The number of untypable strains was higher in the present study than in the experience of Sakazaki et al. (4), and the antigenic types found in the diarrhoeic strains (excepting types 01: K38, 04: K12, 04: K55, 05: K15 and 05: K30) were not usually seen in the environment. A similar pattern has been noted in *Yersinia enterocolitica*, to which *V. parahaemolyticus* is thought to be taxonomically related (1, 3). It is conceivable that unlike *V. cholerae*, *V. parahaemolyticus* is widely distributed in our surroundings, but that nevertheless the risk of infection is limited by the paucity of enteropathogenic serotypes.

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Table 1. Serotypes of *V. parahaemolyticus* strains isolated from different sources in Calcutta

Results	Non-marine fish and water (52 strains)	Human diarrhoea (62 strains)	Marine fish and water (113 strains)
No. typable	17 (32.6 %)	51 (80.2 %)	46 (40.7 %)
No. untypable	35 (67.4 %)	11 (19.8 %)	67 (59.3 %)
Serotypes identified (no. of strains)	01 : K33 (1) <sup>a</sup>	01 : K38 (5)	01 : K38 (1) <sup>b</sup>
	02 : K3 (1) <sup>c</sup>	01 : K56 (5)	02 : K3 (1) <sup>c</sup>
	02 : K28 (4) <sup>c</sup>	03 : K4 (2)	02 : K5 (1) <sup>a</sup>
	05 : K15 (1) <sup>b</sup>	03 : K33 (4)	02 : K8 (1)
	05 : K17 (2) <sup>c</sup>	04 : K8 (1)	02 : K28 (4) <sup>c</sup>
	05 : K47 (2)	04 : K12 (2)	03 : K29 (2)
	05 : K15/17 (1) <sup>d</sup>	04 : K13 (2)	04 : K12 (2) <sup>b</sup>
	08 : K20 (2) <sup>c</sup>	04 : K55 (2)	04 : K29 (2) <sup>a</sup>
	010 : K39 (1) <sup>a</sup>	05 : K12 (1) <sup>a</sup>	04 : K34 (1)
	011 : K22 (1) <sup>a</sup>	05 : K15 (5)	04 : K37 (1) <sup>a</sup>
	011 : K34 (1) <sup>a</sup>	05 : K30 (2)	04 : K55 (1) <sup>b</sup>
		05 : Cal/Ka (12)	05 : K17 (2) <sup>c</sup>
		06 : K46 (1)	05 : K30 (1) <sup>b</sup>
		08 : K21 (2)	05 : K41 (4) <sup>a</sup>
		08 : K22 (2)	05 : K43 (1) <sup>a</sup>
		09 : K23 (2)	06 : K18 (1)
	010 : K19/46 (1) <sup>d</sup>	08 : K20 (3) <sup>c</sup>	
		08 : K38 (1) <sup>a</sup>	
		010 : K9 (1) <sup>a</sup>	
		010 : K24 (3)	
		010 : K26 (5) <sup>a</sup>	
		011 : K38 (3) <sup>a</sup>	
		011 : K54 (1) <sup>a</sup>	
		011 : K38/54 (3) <sup>d</sup>	

<sup>a</sup> Atypical types, i.e., K antigen not linked with specific O group.

<sup>b</sup> Strains resembling human isolates.

<sup>c</sup> Common serotypes in marine and non-marine sources.

<sup>d</sup> Complex types, i.e., occurrence of two K antigens in a single strain.

#### Additional findings

After the preparation of this paper for publication, the authors forwarded the results of continued investigations into *V. parahaemolyticus* serotypes in Calcutta.

The following additional serotypes (number of strains shown in parentheses) were recorded from a second series of isolates of *V. parahaemolyticus* examined after the series described above:

Non-marine strains (35)	Faecal strains (29)	Marine strains (38)
03 : K57 (1)	02 : K3 (2)	01 : K37 (1)
04 : K29 (1)	04 : K9 (3)	01 : K54 (1)
04 : K42 (3)	04 : K11 (1)	03 : K5 (1)
05 : K5 (2)	08 : K20 (2)	011 : K50 (1)
05 : K30 (1)	010 : K19 (1)	
011 : K20 (1)	010 : K24 (1)	

When tested for the Kanagawa phenomenon (1), 82 (90.1%) of the total of 91 human faecal strains examined in both series showed positive results; the serotypes of the 9 negative strains were 03 : K33 (2), 04 : K8 (1), 04 : K12 (1), 04 : K13 (1), 05 : Cal/Ka (1), 06 : K46 (1), and 08 : K20 (2). On the other hand, only 2 of the 238 strains from marine and non-marine sources, belonging to serotype 05 : K47, were Kanagawa-positive. Evidently Kanagawa-positive cultures are readily obtained from stools of patients but not from strains from the Calcutta environment. The question whether Kanagawa-positivity is related to enteropathogenicity has still to be elucidated.

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