

ISOLATIONS OF "GROUP F VIBRIOS" FROM HUMAN STOOLS

Selena Y S Lam
L T Goi

SYNOPSIS

Both anaerogenic and aerogenic strains of Group F vibrios have been recently isolated in Singapore. All strains of *Vibrio fluvialis* were recovered from diarrhoeal stools while a strain of *Vibrio furnissii* was from the stool of an asymptomatic carrier. Biochemical reactions of the *V. furnissii* strain were similar to those reported by Brenner et al. (5) except that it failed to ferment mannose or to grow in the presence of the vibriostatic agent O/129. It was generally susceptible to the routine range of antibiotics tested for enteric pathogens, but resistant to ampicillin. Although the aerogenic *V. furnissii* has been isolated from human diarrhoeal stools, its pathogenicity is doubtful. However, its presence in the stools of healthy carriers should be an important consideration in the study of diarrhoeal diseases.

INTRODUCTION

In 1975, Furniss et al. (1) isolated the first strain of Group F vibrios from the diarrhoeal stool of a Bahrain patient. Subsequently, isolations were reported in Bangladesh and the United States (2, 3). Group EF 6 vibrios described by Huq et al. (2) were found identical to the Group F vibrios of Furniss (1). All human strains were first thought to be anaerogenic except for one strain isolated in Indonesia (3) which demonstrated gas production. In 1981, Lee et al. (4) proposed the name *Vibrio fluvialis* to include both aerogenic and anaerogenic strains of Group F (synonym Group EF 6) vibrios found in the stools of human and animals, and in the environment. According to Sakazaki (5), most human strains isolated in Japan were aerogenic and studies of Brenner et al. (5) during two outbreaks of acute gastro-enteritis among American tourists returning from Japan and Hong Kong confirmed that the aerogenic strains of Group F vibrios belonged to a separate species, and they named the new species *Vibrio furnissii*.

Enteric Bacteriology Laboratory
Pathology Department
Alexandra Hospital
Singapore

Selena Y S Lam, B Sc, M Sc
Bacteriologist

L T Goi
Laboratory Technician

In Singapore, *V. fluvialis* was first isolated from the diarrhoeal stool of a female patient in Changi Hospital in January 1981 (6). She had no fever, abdominal pain or vomiting. Since then, six more isolations were reported in 1981, four in 1982 and three in 1983. All patients were adults. The *Vibrio* strains were generally susceptible to the routine range of antibiotics used except ampicillin (57%), streptomycin (93%) and sulphonamide (93%). In this paper, we describe the isolation of an anaerogenic strain of *V. furnissii* from the stool of a carrier.

CASE REPORT

On 19th March 1984, four persons had food poisoning with acute diarrhoea, abdominal pain and nausea, but no fever. Two of them also experienced vomiting and headache. The stools were not examined as the patients recovered soon after admission to Middleton Hospital. However, epidemiological investigations of the outbreak were traced to chicken rice sold in a food stall. Food samples were later examined and food handlers screened. All test materials were found free from enteric pathogens except for the stool of a 16 year-old male taken on 26th March from which *V. furnissii* was isolated. The patient was asymptomatic at the time of investigation. Subsequently, his stools taken on three consecutive days failed to recover the aerogenic vibrios.

MATERIAL AND METHODS

The stool specimen was cultured on solid media such as eosin methylene blue, thiosulphate citrate bile salts sucrose (TCBS), MacConkey and blood agar and

in enriched selenite F broth and alkaline peptone water added with 2% NaCl (salted-APW). After 37°C incubation overnight, vibrio-like colonies on TCBS and blood agar were identified biochemically. A separate method using Skirrow's medium was employed for the culture of *Campylobacter* species. Antibiotic susceptibility of the *V. furnissii* strain was determined with routine test discs (BBL-Sensi Disc) by the method of Bauer et al. (7). All test media used in the biochemical reactions were added with 1% NaCl and incubated at 37°C for 48 hours unless indicated.

RESULTS

Vibrio-like colonies from the TCBS (sucrose-positive) and blood agar (haemolytic) were identified and found to be similar biochemically as shown in Table I. Most of the reactions conformed to those of *V. furnissii* (5), except that the local strain required three days to dehydrolase arginine. It failed to produce indole or to utilize malonate and tartrate. It did not ferment mannose, rhamnose and cellobiose, nor grow in the presence of the vibriostatic agent O/129. OPNG was positive after one day. The stool was negative for other common enteric pathogens such *Salmonella*, *Shigella*, *Escherichia*, *Yersinia* and *Campylobacter* species.

The *V. furnissii* strain was susceptible to the following antibiotics: kanamycin (30 ug), streptomycin (30 ug), chloramphenicol (30 ug), trimethoprim/sulfamethoxazole (1.25 ug/23.75 ug), tetracycline (30 ug), neomycin (30 ug), sulphonamide (250 ug), gentamicin (10 ug), nalidixic acid (30 ug), carbenicillin (100 ug), cephalothin (30 ug) and Polymyxin B (50 and 300 I.U.). It was resistant to ampicillin (10 ug).

Table I Biochemical characteristics of *V. furnissii* strain isolated from an asymptomatic carrier

Indole	—	Carbohydrate fermentation:	
Methyl red	+ ¹	Glucose	Acid & gas
Voges Proskauer	—	Lactose	—
Citrate (Simmons)	+ ¹	Sucrose	+
Mucate	—	Maltose	+
Malonate	—	Galactose	+
Tartrate (Jordan)	—	Mannose	—
Esculin	—	Arabinose	+
-galactosidase	+ ¹	Rhamnose	—
Oxidase	+	Xylose	—
Nitrate reduction	+	Melibiose	—
Urease (Christensen)	—	Cellobiose	—
Motility	+	Trehalose	+
O/129 sensitivity	—	Raffinose	—
Hydrogen sulphide	—	Mannitol	+
Decarboxylation (Moeller):		Adonitol	—
Lysine	—	Sorbitol	—
Arginine	+ ³	Dulcitol	—
ornithine	—	Inositol	—
Salt tolerance: 0% NaCl	—	Salicin	—
3% NaCl	+ ¹		
7% NaCl	+ ¹		
11% NaCl	—		

(+¹⁻³ = positive between one and three days)

DISCUSSION

Like most vibrios, the *V. furnissii* strain isolated locally was sensitive to the vibriostatic agent O/129. An unusual characteristic was its failure to ferment mannose while the strains reported by Brenner et al. (5) were 100% and the aerogenic Biovar II vibrios of Lee et al. (4) were 74% positive.

The role of *V. fluvialis* as a causative agent of gastroenteritis was first thought to be doubtful until it was associated with diarrhoea outbreaks in Bangladesh and the United States (8). Similarly, although *V. furnissii* has been recovered from human diarrhoeal stools, its pathogenicity is still unknown. However, its presence in the stools of an asymptomatic food handler and a healthy woman in Lima (5) confirmed the existence of human carriers. This finding may provide a significant contribution in the studies of *V. furnissii* and human diarrhoeal diseases.

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