Isolation of *Plesiomonas Shigelloides* in Malaysia

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Introduction

FERGUSON AND HENDERSON, in 1947, reported the isolation of a motile, anaerogenic paracolon organism which they designated as C27, sharing common antigens with **Shigella sonnei.** In Ceylon, Schmid, Velaudapillai and Niles in 1954 reported the study of 12 C27 strains, 4 of which were from domestic animals (sheep, goat, cow and polecat). Bader (1954) described a bacterium, the O antigens of which were related to those of **Shigella sonnei** and assigned the micro-organism to the genus **Pseudomonas** on the basis of its polar monotrichous flagellation. But Ewing and Johnson (1960) suggested that the C27 organism was more closely related to the genus **Aeromonas.**

Habs and Schubert in 1962 (cited by Eddy and Carpenter 1964) suggested that a new genus should be created for this organism and proposed the name **Plesiomonas** (meaning neighbour monad, i.e. to **Aeromonas).**

The epithet shigelloides (shigella-like) was employed by Bader (1954) because the organism possessed common antigens with **Shigella sonnei.** The name **Plesiomonas shigelloides** has been retained despite the finding that only some of the strains have antigens common with shigella organisms, because the specific epithet was validly published according to the Bacteriological Code.

Cooper and Brown in 1968 isolated 38 strains of **Plesiomonas shigelloides** from the faeces of 36 children and 2 adults in Australia and they used the name **Plesiomonas shigelloides.**

The current paper describes the isolation of such an organism from the faeces of a 35-year-old male Chinese tin mine worker, admitted to the medical unit of the University Hospital in Kuala Lumpur.

Clinical features

The patient had a one-week history of a high, dry, remittent fever associated with chills, rigors, nausea and vomiting. For the first four days of his illness, he was constipated but subsequently he passed watery yellow stools once or twice a day.

There was no past history of significance. However, two months previously, his mother had a similar illness which lasted for 10 days.

On physical examination, he was febrile with a temperature of 39.8° C, was dehydrated and mildly jaundiced. His blood pressure was 140/70 mm.Hg.

The abdomen was mildly distended with tenderness over the left paraumbilical region. The liver was enlarged to 6 cms. and the spleen to 1½ cms. below the right and left costal margins.

There was no significant lymphadenopathy. The other systems were normal. His haemoglobin was 14.4 gm/100 ml., white blood count 6,600/ml., with a differential count of neutrophils 35%, lymphocytes 59%, monocytes 4%, atypical lymphocytes 2%. His serum bilirubin was 2.5 mg/100 ml. with 0.6 mg. conjugated and 1.9 mg. unconjugated. The urine did not contain any bile or urobilinogen.

In view of the above findings, a provisional

diagnosis of typhoid fever was made, and the patient started on chloramphenicol therapy.

Methods of Isolation

The patient's faeces were cultured bacteriologically 7 times during his stay in hospital. The faeces were plated on MacConkey agar and deoxycholate-citrate agar and incubated at 37°C for 18 hours. Part of the specimen of faeces was also cultured in selenite F medium for 18 hours and then was plated onto MacConkey and deoxycholate-citrate agar.

All cultures were examined for non-lactose fermenting colonies and these were subcultured onto Kligler iron agar slopes which were incubated at 37°C for 18 hours. On inspection after incubation, the results were an acid butt and alkaline slope with no gas or hydrogen sulphide production – a reaction similar to that given by the shigella group of organisms. Accordingly, a slide agglutination with **Shigella sonnei** antiserum gave a very strongly positive reaction. But subsequent biochemical reactions did not confirm the organism as a **Shigella sonnei.** These suggested the organism to be **Plesiomonas shigelloides,** and this was confirmed by Dr. Cooper in Adelaide. The organism was isolated only from the first two of the 7 specimens of faeces examined. No organism of the dysentery or enteric group was isolated.

Bacteriological Features

Plesiomonas shigelloides was a motile gram negative rod which formed smooth, colourless and convex colonies, 1–1.5 mm. in diameter with entire edge on deoxycholate-citrate medium after incubation for 24 hours. It was a facultative anaerobe growing well both aerobically and anaerobically on blood agar as well as on nutrient agar. In Kligler iron agar, the reaction given was similar to that of shigella organisms – acid butt, alkaline slope with no gas or hydrogen sulphide being produced. The following table gives the bacteriological features of the organism.

Serology

The organism gave a strong positive slide agglutination reaction and agglutinated in dilutions of up to 320 in a tube test with **Shigella sonnei** phase I & II antiserum (Burroughs Wellcome).

Bacteriological features of Plesiomonas shigelloides

TEST	RESULTS
Motility	+
Catalase	+
Cytochrome oxidase	+
Gas in glucose	–
Acid in glucose (1 day)	+
Acid in lactose (1 day)	–
Acid in lactose (3 days)	+
Acid in sucrose (1 day)	–
Acid in maltose (1 day)	+
Acid in mannitol (1 day)	–
Acid in dulcitol (1 day)	–
Acid in inositol (1 day)	+
Acid in salicin (1 day)	–
Acid in xylose (1 day)	–
H2S (Kligler)	–
Indole	+
Methyl Red	+
Voges Proskauer	–
Citrate utilisation	–
Urease	–
Malonate utilisation	–
Phenylalamine deaminase	–
Lysine decarboxylase	+
Arginine decarboxylase	+
ONPG reaction	+

Antibiotic Sensitivity

Sensitivity to antibiotics was tested on plates of Oxoid sensitivity agar using MAST antibiotic discs. The organism was found to be sensitive to ampicillin, cephaloridin, streptomycin, tetracycline, chloramphenicol, neomycin, kanamycin and trimethoprim.

Discussion

Cooper and Brown, in their paper, discussed the similarities as well as the difference between **Plesiomonas shigelloides** and **Aeromonas.** A characteristic feature of this organism is that it sometimes has **Shigella sonnei** antigens. Not all the isolates possess this feature, but some of them may have other shigella antigens.

In Japan, Honi, Hayashi, Maeshima, Kigawa, Miyasato, Yoneda and Hagishara in 1966 (cited by Cooper and Brown 1968) described 10 strains which had

Shigella dysenteriae type 7 antigen. Cooper and Brown in Australia found that only 4 of their 38 strains had **Shigella sonnei** antigens and one had **Shigella felexneri** type 6 antigens. Eddy and Carpenter (1964) found that out of their 21 strains, only 12 possessed the **Shigella sonnei** phase I antigen.

Thus the majority of the strains do not have any shigella antigens. Therefore an organism giving shigella-like reaction in Kligler iron agar and not agglutinated by shigella antisera must be tested for **Plesiomonas shigelloides**. A presumptive identification can be made if it is also motile, oxidase positive and mannitol negative.

The question that cannot be answered categorically is whether this organism gives rise to a dysentery-like illness.

Our patient was provisionally diagnosed as a case of enteric fever but was never confirmed bacteriologically or serologically. However, he made an uneventful recovery after chloromycetin therapy, and no **Plesiomonas shigelloides** or dysentery or enteric type of organisms were isolated subsequently.

No other cause of his illness was elucidated. Blood cultures, 4 specimens, were negative; Widal and Weil-Felix titres were not significant.

Are we entitled to say that the cause of his illness was **Plesiomonas shigelloides**? Cooper and Brown isolated this organism from infants suffering from mild as well as severe diarrhoea. They believe that this organism can be the cause of enteritis especially in infants, although the organism may be found in the absence of enteritis. Some of the C27 strains of Eddy and Carpenter (1964) also came from human patients, mainly with diarrhoea.

Other workers in Ceylon and in Japan have isolated the organism from domestic animals, healthy adults as well as from children and adults suffering from diarrhoea. This is reminiscent of the food poisoning type of Salmonella which can be isolated from a variety of domestic animals, reptiles, healthy human beings as well as from people suffering from gastro-enteritis.

It may well be, as Cooper and Brown pointed out, that possession of shigella antigens may confer on them the ability to produce illness. It is hoped that as more medical microbiologists become aware of this organism, more evidence will be brought forward for the pathogenicity or otherwise of **Plesiomonas shigelloides.**

It is believed that this is the first reported isolate in Malaysia and further work on it is in progress.

Summary

Plesiomonas shigelloides was isolated from the bowel of a patient with pyrexia and diarrhoea. The morphology and biochemical reactions of the organism are described and its significance discussed.

Addendum

After preparing this manuscript, a further 8 strains of **Plesiomonas shigelloides** were isolated from patients with diarrhoea.

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