

Changing Trends of Antibiotic Susceptibility Pattern among *Vibrio cholerae* O1 Serovars from Sporadic Cases in Coastal Karnataka, South India

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ABSTRACT

Changing patterns of antibiotic sensitivity of *V. cholerae* is a major concern as Multiple-Antibiotic-Resistant (MAR) *V. cholerae* isolates are being reported from various parts of the world. The study was conducted to analyze the prevalence and antibiotic resistance pattern of *V. cholerae* O1. Samples were processed according to the standard guidelines. Sporadic outbreaks occurred in June 2004 and March 2010. Antibiotic resistance was seen in the strains isolated. A large number of cholera epidemics have been associated with MAR strains of *V. cholerae*. This makes it imperative that all isolates be constantly subjected to susceptibility testing and resistance patterns to each antibiotic be monitored.

Keywords: *Aeromonas hydrophila*, *Vibrio cholerae* Inaba, *Vibrio cholerae* Ogawa, Tetracycline

INTRODUCTION

Vibrio cholerae is one of the major etiological agents causing watery diarrhoea in almost all parts of the society, especially in the lower socio economic strata of developing countries like India. Sporadic or epidemic cases of cholera are reported frequently from both adults and children alike. Epidemics of cholera caused by toxigenic *V.cholerae* O1 and *V.cholerae* O139 represent a major public health problem¹ for India with frequent epidemic potential as an important cause of morbidity and mortality. The use of antimicrobial agents in cholera is generally accepted as a method of reducing the duration and volume of diarrhea as well as decreasing the period of *Vibrio cholerae* excretion in stool.²

MATERIALS AND METHODS

Fecal specimens from patients with acute or watery diarrhea during the sporadic outbreak were received for microscopy and culture of routine enteric pathogens at

Diagnostic Microbiology Laboratory, during June 2004 and March 2010. Sixty and 92 stool specimens were collected before antibiotic administration and processed for identification of various pathogens respectively. No transport medium was employed as fresh specimen was received in the laboratory within 30 minutes and processed immediately without any further delay. Routine stool microscopy of saline and iodine preparations was examined for intestinal parasites. A battery of culture media - Alkaline Peptone Water, Selenite Feces broth, Blood agar, Mac Conkey agar, Thiosulphate Citrate Bile salt Sucrose agar (TCBS) were used for the isolation of bacterial pathogens. Bacteriological analysis was done by standard laboratory techniques.^{3,4} *Vibrio cholera* was serotyped by slide agglutination test using specific antisera (Difco, USA and NICED, Kolkata). The antimicrobial susceptibility testing was performed for *V. cholerae* by using the Kirby Bauer disc diffusion technique using commercially available discs (Hi-media). The antibiotics used were ampicillin (AMP 10 µg), chloramphenicol (CHL 30 µg), tetracycline (TET 30 µg), cotrimoxazole (SXT 25 µg), ciprofloxacin (CIP 5 µg), nalidixic acid (NAL 30 µg) and gentamicin (GEN 10 µg). The current CLSI guidelines were followed for interpretation of the results and *E.coli* ATCC 25922 was used as the control strain.

Patients' clinico-demographic details which include age,

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sex, ward / OPD, presenting clinical history and underlying illness were noted wherever applicable. Institutional Ethical Committee approval was obtained.

RESULTS

An outbreak of cholera occurred in the coastal town, in the month of June, 2004. A total of 100 patients were admitted to our tertiary care hospital with the presentation of acute watery diarrhea with moderate to severe dehydration. Out of the total patients, 60 stool samples were received in the laboratory for the identification of the causative agent, of which 36 (60%) stool samples were found to be positive for *V.cholerae*. Thirty three (91.6%) samples were positive for the biotype El Tor serotype Ogawa and 3 (8.3%) samples were positive for serotype Inaba. Another sporadic outbreak occurred during March to June, 2010. A total of 92 stool samples were processed in the laboratory, of which 11 samples (11.95%) were found to be positive as *Vibrio cholerae* O1 biotype El Tor serotype Ogawa.

The analysis of antimicrobial resistance in 2004 and 2010 as shown in Figure I makes it evident that there is a change in the resistance pattern of the isolates. In 2010 there was increased resistance of the *Vibrio cholerae* isolates towards TET, SXT & AMP, however there was a decreased resistance towards CHL, CIP & GEN.

DISCUSSION

V.cholerae O1 Inaba existed for a brief period in 2004 and was replaced by *V. cholerae* O1 Ogawa in the subsequent years. *Vibrio cholerae* Ogawa was the

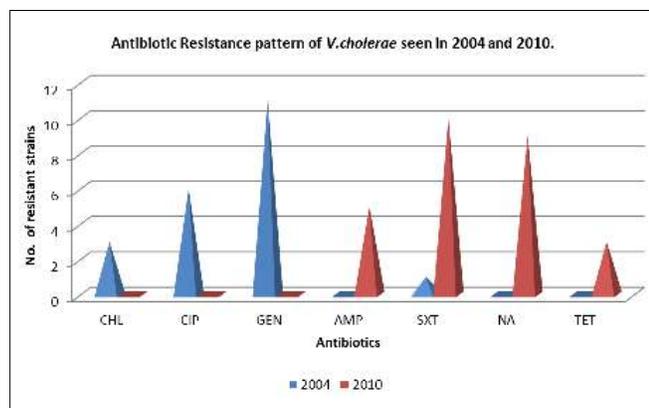


Fig. 1: Graphical representation of antibiotic resistance pattern of *V.cholerae* O1 seen in 2004 and 2010.

predominant isolate in 2004 as well as in 2010. During the last decade, the dominance of Ogawa belonging to the O1 serotype has seen a shift to Inaba in the last few years of the decade as cases reported from Chandigarh, Bangladesh and Vellore.^{2,5,6}

Changing patterns of antibiotic sensitivity of *V.cholerae* is a major concern as multiple antibiotic resistant *V.cholerae* isolates are being reported from various parts of the world.^{5,7-9} In the present correspondence, we report the appearance of tetracycline resistance in *V.cholerae* O1 biotype El Tor serovar Ogawa isolated in this coastal region of India. The observation has epidemiological and clinical importance as tetracycline has been the main stay of antibiotic treatment for cholera. In India, tetracycline resistance has been reported in Inaba serovar but sparsely reported in Ogawa serovar.^{5,6}

It is important to note that tetracycline-resistant *V.cholerae* O1 Ogawa isolates are being reported from Madagar, Bangladesh, Tanzania, Zaire, Latin America and India (Southern and Eastern regions). The pattern of shift in antibiotic resistance indicates an enhanced mobility in genetic elements, which confer resistance to antibiotics. Recently genetic elements, such as a class I integrin and SXT constin carrying tetracycline-resistant genes, tetG and tetA respectively have also been reported to be associated with the spread of genetic determinants of resistance to antimicrobial agents.^{9,10}

The extensive use of tetracycline / doxycycline might have led to the rapid emergence and spread of tetracycline-resistant isolates of *Vibrio*. The molecular mechanism of tetracycline resistance in *Vibrio* must be studied and the transfer of such resistant genes has to be epidemiologically monitored.

To conclude, a large number of cholera epidemics have been associated with multiple-antibiotic-resistant (MAR) strains of *V.cholerae*.¹¹ A sudden increase in antibiotic resistance as reported here cannot be ignored; future epidemics in the MAR (Multiple Antibiotic Resistance) serotypes may be imminent. This makes it imperative that all isolates be constantly subjected to susceptibility testing and resistance patterns to each antibiotic to be monitored. Thus antibiotics should be judiciously used in the management of diarrheal diseases. The importance of reporting all cases should be emphasized, with the ultimate goal being a thorough

understanding of the constantly changing and obscure epidemiology and resistance patterns of *V. cholera*.

Conflict of Interest: None

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