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Influence of hydrologic and anthropogenic factors on the abundance variability of enteropathogens in the Ganges estuary, a cholera endemic region



Prasenjit Batabyal ^a, Marc H. Einsporn ^{b,*}, Subham Mookerjee ^a, Anup Palit ^a, Sucharit B. Neogi ^{c,d}, Gopinath B. Nair ^{a,f}, Rubén J. Lara ^{b,e}

- ^a National Institute of Cholera Enteric Diseases (ICMR), 700010 Kolkata, India
- ^b Leibniz Center for Marine Tropical Ecology (ZMT), 28359 Bremen, Germany
- ^c International Centre for Diarrheal Disease Research, Bangladesh (ICDDR,B), Mohakhali, Dhaka 1212, Bangladesh
- ^d Graduate School of Life & Environmental Sciences, Osaka Prefecture University, Sakai, Osaka 599-8531, Japan
- ^e Instituto Argentino de Oceanografía, 8000 Bahía Blanca, Argentina
- f Translational Health Science and Technology Institute, Udyog Vihar, Gurgaon-122016, Haryana, India

HIGHLIGHTS

- · Aquatic enteropathogen dynamics with respect to variations between winter and monsoon
- · Synchronous water and sediment sampling from two sites of the River Hooghly
- Tidal and seasonal variation of physico-chemistry correlated to bacterial abundance
- · Indication of bentho-pelagic coupling of Vibrio dynamics

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ABSTRACT

This study deals with the influence of water physico-chemical properties, tides, rainfall and fecal pollution on the abundance of enteropathogens in a main distributary of the Ganges, in the endemic cholera belt of West Bengal. Between January and June 2011, water and sediments were sampled from two sites of the Hooghly River by Kolkata and Diamond Harbour. Counts of cultivable *Vibrio* (CVC, from $\sim 10^2$ to $\sim 10^5$ CFU/L) and total bacteria (TBC, from $\sim 10^5$ to $\sim 10^9$ CFU/L) increased with water temperature (17 °C to 37 °C). A combination of variations in tidal height, salinity and turbidity had a distinct influence on CVC, TBC and coliform counts. At Diamond Harbour, a salinity increase from 0.6 to 7.9 was accompanied by a 1000-fold amplification of initial CVC $\sim 10^2$ CFU/L, whereas higher prevalence of coliforms in Kolkata was related to greater disposal of untreated sewage into the river. Turbidity-dependent variation of CVC was noteworthy, particularly at Diamond Harbour, where CVC in intertidal surface sediments showed an analogous trend as in surface waters, suggesting bentho-pelagic coupling of *Vibrio* dynamics. Besides the influence of salinity variation with tidal cycles, sediment re-suspension from tidal flats can play a role on *Vibrio* abundance in aquatic ecosystems.

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1. Introduction

Diarrheal diseases associated with floods and droughts are common in East, South and Southeast Asia and are expected to increase due to changes in the hydrological cycle (Lipp et al., 2002; IPCC, 2007). Yet, our knowledge on the influence of environmental factors on the regulation of different pathogenic enteric bacteria in estuarine environments

E-mail address: marc.einsporn@zmt-bremen.de (M.H. Einsporn).

is still limited (Lara et al., 2011). Particularly for low-lying coasts in tropical regions, there is a need for comprehensive knowledge on the links between hydrology and the ecology of disease causing agents (Wolanski et al., 2004).

Vibrio cholerae O1 and O139 are the causative agent of cholera, whereas other vibrios (e.g., V. parahaemolyticus, V. vulnificus, V. mimicus, etc.) are responsible for diarrhea, gastroenteritis, necrotizing fasciitis, various further skin diseases and septicemia affecting humans worldwide (Thompson et al., 2004). Cholera is an important cause of morbidity and mortality in many developing countries in Asia, Africa and Latin America due to lack of safe water supply and poor hygienic practices (Colwell, 1996), together with the resulting massive use of untreated,

^{*} Corresponding author at: Leibniz Center for Marine Tropical Ecology (ZMT), Fahrenheitstrasse 6, D - 28359 Bremen, Germany. Tel.: +49 421 238 00 46; fax: +49 421 238 00 30.

often highly polluted surface waters for human consumption. Even in the present decade, hundred thousands of people suffer from cholera every year, e.g., during 2010, there were approx. 318,000 reported cases of cholera, with 7543 deaths, though the actual death toll could have been as high as 120,000 (WHO, 2012).

Most cholera pandemics started in the Ganges-Brahmaputra delta, considered as the homeland for cholera since ancient times (Drasar and Forrest, 1996). Vibrio cholerae is an endemic inhabitant of tropical estuaries, and cholera epidemics can be related to changes in the physico-chemical properties of water, its main transmission vehicle (Singleton et al., 1982). Further, the input of untreated sewage from cities into the riverine environment increases the risk of pathogen transmission between humans using those waters for bathing, cooking or drinking. But Vibrio cholerae can be further concentrated in the aquatic environment. For example, zooplankton, fish or shrimps can host the bacteria and represent an additional threat to human health consuming these organisms (Colwell, 1996). Although a few studies were conducted to identify the favorable physico-chemical conditions for Vibrio survivability and growth (Chatterjee and Gupta, 1959, Miller et al., 1982, Dunlap, 2009), the microbial dynamics of this large aquatic ecosystem, characterized by high loads of organic matter and suspended sediments, is poorly understood.

The waters of the Ganges are used by millions of people for an enormous variety of personal, societal and industrial purposes; especially near the megacity of Kolkata, a cholera endemic region. The present study deals with links between variations in hydrologic conditions, water biogeochemistry and *Vibrio* abundance in the Hooghly River, a main distributary of the Ganges. Furthermore, under consideration of microbiological indicators of inputs from Kolkata, this approach aims to discriminate between hydrologic and anthropogenic drivers of change of enteric pathogen abundance, as a contribution to the understanding of cholera dynamics in West Bengal.

2. Methods

2.1. Study sites

The Hooghly River originates at the junction of the rivers Bhargirathi, Jalangi and Mathabhanga. Since simultaneous samplings at several locations along the estuarine gradient would have involved an unaffordable logistic effort, two stations with contrasting environmental settings, Howrah Bridge and Diamond Harbour, were chosen and sampled during winter and summer, from January to June 2011 (Fig. 1).

The Howrah Bridge site (HB) is within an urban setting connecting the City of Kolkata and the township of Howrah (approx. 15 million inhabitants). Although 130 km away from the sea mouth of the Hooghly River, this site has an estuarine condition due to a significant tidal oscillation of ~3 m. Upstream of HB, the river flows through the densely populated region in south-east West Bengal, as well as through many areas of agricultural and aquaculture use. The river water is accessed for drinking, washing, bathing, cleaning utensils and for many religious rituals. Mostly untreated sewage from both sides of the river is disposed off into its waters.

Diamond Harbour (DH) hosts an important commercial port. It is surrounded by a rural hinterland and located about 50 km downstream of HB. Being closer to the estuary mouth, the river is ca. five times wider than in Kolkata and has a higher turbulence. Also the tidal influence is larger, with higher seawater intrusion during the flood, while large mud flats get exposed at low tide. The human impact on the estuarine environment is less at this site due to lower population density, strong water currents and exchange with the sea.

2.2. Sampling of surface waters and sediments

Sampling was carried out every three weeks, in order to embrace the effect of hydrological changes due to the different tidal range in alternating spring and neap tides, as well as during increasing and decreasing moon phases. Synchronized sampling was undertaken at both sites. At each sampling day, 5 to 7 samples were collected at regular intervals (every 2 h) covering a 12 – 14 h period of a tidal cycle including low and high tides. During each sample collection, three to four subsamples of surface water from the midstream were taken with a metal bucket and pooled together in a sterile 10 L container. Subsequently, the samples were shaken and divided into 1 L aliquots. Additionally, surface sediment samples were taken from a tidal flat at Diamond Harbour. At low tide, 10 sub-samples of 3 cm³ of mud were taken with syringes from a surface of 1 m² and pooled together in a sterile glass flask. After collection, water and mud samples were put into icechilled, lightproof boxes and transported to the laboratory for further processing and analysis within 12 h of collection.

2.3. Physico-chemical and hydrological parameters

Immediately after sample collection, temperature, pH, conductivity and salinity were measured with a Multi-meter (pH/Cond 340i WTW, Weilheim, Germany). Turbidity was measured by a portable turbidimeter TD-100 (Eurotech, Singapore) and expressed as Nephelometric

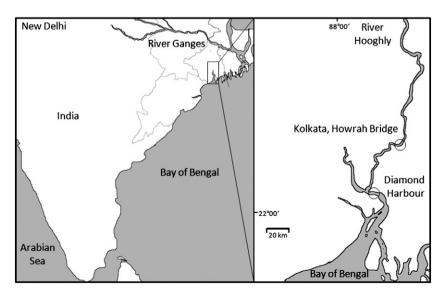


Fig. 1. Map of sampling sites in the estuary of the Hooghly River. Both sites, Howrah Bridge (HB, 22° 35′ 6″ N, 88° 20′ 49″ E) and Diamond Harbour (DH, 22° 11′ 37″ N, 88° 10′ 48″ E).

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