Prevalence of rotavirus infections and strain types detected among under-five years children presenting with diarrhoea at MCH clinics in Mwanza City, Tanzania

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Abstract: The prevalence of rotavirus infections and strain types was determined in 805 underfive children presenting with diarrhoea at selected MCH clinics in Mwanza City, Tanzania. The study was conducted in February-May 2001 as an initial stage of conducting detailed epidemiological studies on rotavirus infections among underfive children presenting with diarrhoea in the Lake Victoria basin. Stool specimens were collected from underfive children presenting with diarrhoea at Makongoro and Hurumia Watoto MCH clinics within the City. Rotavirus antigen was detected in 79 (9.8%) of all stool specimens collected. The majority of rotavirus strains (93%) were of the long electrophoretic type. VP6 subgroup II viruses were more common than subgroup I (70% *versus* 5%). VP7 serotype G1 was the most common (61%) compared to other serotypes. Rotavirus infection was detected in 9.8% of stool specimens collected from under five children presenting with diarrhoea in Mwanza City.

Introduction

It is estimated that approximately 2.4-3.3 million deaths due to diarrhoea in underfives occur annually in developing countries where the disease is a major public health problem (*1*). Rotavirus infections have been shown to be very common in children under one-year-old with diarrhoea, and the highest incidence being in those aged 6-11 months (*2*). However, in Tanzania the proportion of diarrhoea cases associated with rotavirus infection is poorly understood though the disease is a major cause of morbidity and mortality especially in underfive children.

The 1995/96 annual report of the Department of Paediatrics at Bugando Medical Centre (BMC), showed that 15% (500/3358) of admissions were due to diarrhoea, and 13.7% (51/372) of deaths were associated with diarrhoea. These figures may underestimate the true magnitude of diarrhoea, because BMC is one of the four apex hospitals in the country, thus only few cases of diarrhoea rich this level, and most most most of the cases are either seen at the dispensary or health centre levels.

Strategies to reduce diarrhoea morbidity and mortality in Tanzania have included improved case management and improved availability of water and personal and domestic hygiene. It has been reported that improved case management, water availability and personal and domestic hygiene were associated with marked reduction in mortality (*3*), and prevalence, incidence, morbidity and mortality due to diarrhoea (*4*).

Rotavirus vaccines have been developed and shown to be protective (5). Therefore, application of the vaccines could compliment other diarrhoea disease control strategies. However, development and application of appropriate rotavirus vaccines requires an understanding of the magnitude of diarrhoea associated with rotavirus infection, the age group most affected, the severity of diarrhoea associated with rotavirus infections and the strain types circulating in an area. The study which is reported here attempted to determine the prevalence of rotavirus infections and strain types circulating among underfive children presenting with diarrhoea in Mwanza City as an initial step into understanding the epidemiology of rotavirus infections in the country.

Materials and methods

Study population and study sites

Mwanza City is situated on the southern shore of Lake Victoria. The City population estimated at 400,000 has increased dramatically recently following introduction of commercial fishing and mining in areas close to the City. This has resulted in unplanned human settlement and hence unsafe disposal of waste products. This condition could result in increased diarrhoea cases.

Stool specimens collection

Children presenting with diarrhoea at Makongoro and Hurumia Watoto MCH clinics were requested to participate in the study. Stool specimens were collected from February to May 2001.

Caretakers of children presenting with diarrhoea at these clinics were informed about the study and were requested for their children to participate by providing stool specimens. Caretakers who consented went through the routine MCH clinic procedures and at the end were provided with stool containers for specimen collection. The management of diarrhoea followed standard national guidelines. Stool specimens submitted were kept in a cool box at the clinic until transported to the National Institute for Medical Research (NIMR) laboratory in Mwanza at the end of the clinic session.

Laboratory procedures

Stool specimens were immediately examined microscopically at the clinic for intestinal parasites and the results were given back to the clinic staff as they become available. At the NIMR laboratory stool specimens were kept at 4°C until tested. ELISA (IDEIA, DAKO) was used to screen all stool specimens for rotavirus antigen. Stool specimens reactive for rotavirus antigens were subjected to Polyacrylamide Gel Electrophoresis (PAGE) for electrophoretype determination. Furthermore, rotavirus antigen reactive stool specimens were tested for presence of subgroup and serogroup specific antigens by ELISA using monoclonal antibodies. Both PAGE and strain grouping and serotyping was done at the Medical University of South Africa.

Results

A total of 805 stool specimens were collected from under-five children presenting with diarrhoea. The age of children who provided stool specimens ranged from 1 to 48 months. Three hundred and fifty three (43.8%) of the 805 stool specimens submitted had pus cells. Hookworm was detected in 1.1% (9/805) of stool specimens, 2.3% (18/805) had *Giardia lamblia*, 10.2% (82/805) had *Entamoeba histolytica* and 2.7% (22/805) had *Ascaris lumbricoides*.

Rotavirus antigen was detected in 9.7% (79/805) of stool specimens collected. PAGE analysis of the rotavirus RNA showed presence of both profiles though those belonging to the long type were the majority (93%). Rotavirus strains belonging to the VP6 subgroup II were more common than subgroup I (70% *versus* 5%). Several serotypes of VP7 were detected, being G1 in 61%, G2 in 2% and G1/G2 mosaic strains in 7% of stool specimens. Serotype G3 and G4 were not detected.

Discussion

The study findings suggest that the water source for Mwanza City is contaminated with faecal matters and the process of treating water is not effective enough to eliminate *Giardia lambria* and *E. histolytica* cysts. Detection of *G. lambria* and *E. histolytica* trophozoites and cysts in these young children is of great concern because one of the strategies to reduce diarrhoea associated morbidity and mortality is through improved water availability both in terms of quality and quantity (*3, 4*). Thus diarrhoea will continue to be a major public health problem unless the problem of poor water quality is addressed.

We used direct microscopy for examination of stool specimens for intestinal parasites at the clinics for immediate case management. This could have resulted into underestimation of the magnitude of parasite infections in these children. A concentration method is more sensitive than direct preparations in detecting parasite eggs and cyst but this cannot be done at the clinic. Then, this would have required taking specimens to NIMR laboratory for processing and hence a delay in providing results to the clinic.

The prevalence of rotavirus infection in this population was low and could be due to the period the specimens were collected. We only collected our specimens from February to May. This may affect the observed prevalence if this period was not the time when highest transmission of rotavirus occurs and hence high prevalence of rotavirus associated diarrhoea. Rotavirus infections have been reported elsewhere to be the most common cause of acute gastroenteritis especially in children aged less than two years (*6*). It has been reported that rotavirus associated gastroenteritis shows seasonality though transmission may occur throughout the year (*7,8*).

Alternatively, the delay in screening of the specimens for rotavirus antigens due to delay in receiving the ELISA kits may have contributed to the low prevalence observed in this study. Stool specimens were kept at 4°C before were tested for rotavirus antigens. A longer storage time at this temperature may have resulted into natural breakdown of antigen molecules and hence reduced the number of antigen epitopes to be detected. This could have affected most those specimens with a low number of virus particles.

Water source in Mwanza City seems to be contaminated with faecal matter and the process of water treatment is not effective or contamination occurs along

the distribution system. Therefore, water quality need to be improved in order to reduce diarrhoea disease burden in the population of Mwanza City. Rotavirus could be an important cause of diarrhoea in under-five children in this area. There is a need to conduct further studies to determine the actual magnitude of rotavirus associated diarrhoea, the strain types, seasonality and the age of children most affected with these infections in the context of the Lake Victoria basin. The information will be very useful in implementing rotavirus associated diarrhoea control through a vaccination programme.

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