JOURNAL OF PURE AND APPLIED MICROBIOLOGY, April 2010. Vol. 4(1), p. 237-241

Microbiological Study of Gastroenteritis During Summer

Suchismita Patra[1], Sukantibala Mohapatra[1], Dipti Sunder Mohanty[2] and Pravas Ranjan Misra[2]

[1]Department of Microbiology, Centre for Post Graduate Studies, Orissa University of Agriculture and Technology, Bhubaneswar, India.
[2]Asian Institute of Public Health, Nayapalli, Bhubaneswar, India.

(Received: 12 Septemebr 2009; accepted: 24 Octoebr 2010)

Acute gastroenteritis is one of the leading causes of childhood morbidity and mortality in the developing countries. Hence the present study was undertaken to determine the prevalence of bacterial as well as viral agents associated with gastroenteritis in children, their isolation and antibiogram of the bacterial isolates as a prospective for treatment. Children of age 1 month to 11 year with a history of gastroenteritis were studied. Total 180 subjects with an episode of these ailments were included in this study. Simultaneously 30 children of the same age group without clinical symptoms of gastroenteritis were included as control in this investigation. Out of 180 sample analyzed, 126(70%) were positive only for bacterial agents whereas 54 (30%) samples were found positive for both bacterial as well as viral agents. *E. coli* (180 ;18.09%), *Enterococcus* spp.(162;27.14%), *Clostridium* spp.(63 ;10.45%) were predominantly isolated and identified. Antibiotic sensitivity test of the identified gram positive cocci were found sensitive to glycopeptides (33.3%),aminoglycosides (33.3%), pencillins(33.4%) and mostly gram negative bacillus were sensitive to carbapenems(47%), cephalosporins(23%) and flouroquinolones(19%). It was concluded that bacteria as well as viruses were commonly associated with childhood gastroenteritis. There is a need for further detailed studies on the molecular characterization of rotaviruses which would have important implications in vaccine evaluation programme.

Key words: Bacterial, Viral agents, Gastroenteritis, Antibiotics.

Gastroenteritis is a global problem which is recorded as the leading cause of death and illness among children in the developing countries. Children constitute a major proportion of the global population today. It has been reported that 10% of 5.8 billion people living in the world are children under 5 years of age.[1]Gastroenteritis is a cause of significant morbidity and mortality of children younger than 5 years of age in the Indian subcontinent. [2] It is responsible directly or indirectly for 3 million death every year i.e. statistically 1 in every 10 seconds.[3] It is estimated that annually some 1.8 million children die from the direct effect Of diarrheal disease. Out of approximate 6,00,000 annual death due to rotavirus worldwide more than 150, 000 occur in India . It was also observed that 20 to 30 percent hospitalized cases of diarrhea are due to rotavlrus m our country.

* To whom all correspondence should be addressed.

In the state of Orissa socio-economically back ward classes of tribal population frequently receive the epidemic of diarrheal diseases, accounting high morbidity and mortality .However, there is paucity of recent reports on various epidemiological and bacteriological profiles of acute gastrointestinal disorder in the state. Lack of access to safe drinking water and adequate sanitation are the other under laying factors behind child death . For death, children under 5 years of age, diarrhea accounts the higher than the others . The single most important factors for reducing the prevalence rate and case fatality of major infant and childhood disease are improvement of sanitation, antenatal and intranatal care.

MATERIAL AND METHODS

The present investigation is a prospective case control study conducted during summer, at Asian Institute of Public Health, Bhubaneswasr. Children in the age group of (1month to 11 Years) admitted with acute gastroenteritis during the study period were enrolled in the study, as cases. Permission to collect samples were obtained from the authority of the Capital Hospital , Unit-6, Bhubaneswar and the parents respectively. For both the cases and controls, a detailed history was recorded to elicit various potential risk factors.

A detailed history of relevant symptoms like fever , cough, cold, abdominal cramps, frequency of diarrhea was taken . Socio economic status of the patients were noted and grouped accordingly as Lower income group (LIG), Middle income group (MIG), Higher income group (HIG). Histories of breast feeding , nutritional status, age, sex, and antibiotics they used were obtained from the patients .Controls included in the study were healthy children between 1 month - 11 year were normal sibling of admitted children.

Freshly voided stool samples were collected in sterile container as well as rectal swabs were also collected and carried in ice box having cary- blair transport medium to the labouratory for microbiological analysis. 108 stool samples were collected from cases. For comparative bacteriological study 30 approximately normal stool samples were included. Viral etiological agents were dected by immunuchromatography technique using VIKIA R rota -adeno test device (biomerieux,France). For isolation of bacterial etiological agents, the samples were inoculated into Blood agar, Macconkey agar (for aerobic isolation), Enterococceal agar (for microaerophilic isolation), and Rogosa agar and Anaerobic agar (anaerobic isolation). Then isolated colonies were grouped by gram staining , oxidase test as well as catalase test were done. Different API strips (API-Staph,API-Strep, API-20A, API-20 E, API-20 NE) (biomerieux , France) were used for identification of isolated colonies ,serotyping of the identified *E. coli* strains were done. Antibiotic senSitivIty test of the identified bacterial isolates were done by using Kirby and Bauer (1966) disc diffusion technique.[6]

RESULTS

Over the period of 6 months, 180 children infected with gastroenteritis in Capital hospital were studied. In this investigation, 108 gastroenteritis cases were analysed. Male

Table 1. Sociodemographic variable in gastroenteritis cases

Parameters	No of patients	Percentage
1.Sex of the patients		
Male	117	65
Female	63	35
2.Age of the patients		
>1 yr	87%	48
1-3 yr	51%	28
4-6 yr	24%	14
7-11 yr	18	10
3. Socio-economic status		
LIG	90	50
MIG	75	41.5
RIG	15	8.5
4. Feeding habits		
Mother's milk	24	13.33
Mother's milk with commercial milk	57	31.67
Commercial milk	99	55
5. Water sorces		
Pipe-borne	57	32
Well	123	68
Borehole	0	0

preponderance was found in gastroenteritis cases than females and majority of children were infants in their age distribution .Also, patients, which belong to LIG, category were infected more in comparision with MIG and RIG. When other sociodemographic variables were studied, there were significantly higher number of infected children, fed with commercial milk .The children whose families use well water are infected more than others. Among the etiological agent studied, infection by bacteria was higher than the viral agent. Among the viral agent rotavirus accounts more (23.33%) than adenovirus(6.67%). The bacteria isolated were *E. coli* (108,18.09%), *Salmonella* spp. (12,2.01%), *E. aerogen* (9 ; 1.51%), *F. oryzihabitans*(15,2.51 %), *K. pneumoniae* (15,2.51 %), *Citrobacter* spp. (12,2.01 %), *E. vulneris* (12,2.01%) and *Staph.* spp. (102,16.92 %) isolated aerobically, Microaerophically *Enterococcus* spp. (162, 27.14%) and anaerobically *Lactobacillus* spp. (54, 9.05%), *Clostridium* spp.(63, 10.45%), *Eubacterium* spp. (21,3.51 %) and *Fusarium spp.*(12, 2.01 %) in Table 3. On comparison with the control in gastroenteritis cases gram negative rods were high in percentage than control group.

Table 2. Prevalence of bacterial and viral gastroenteritis

Organism	No.	Percentage
1. Bacteria	126	70
2. Bacteria+virus	54	30
3. Viral agents		
Rotavirus	42	23.33
Adenovirus	12	6.67

Table 3. The different types of bacteria isolated from gastroenteritis

Aerobic	No (Percentage)	Microaerophilic	No (Percentage)	Anaerobic	No (Percentage)
E.coli	108(18.09%)	*Enterococcus* spp.	162(27.14%)	*Lactobacillus* spp.	54(9.05%)
Salmonella spp.	12(2.01%)			*Clostridium* spp.	63 (10.45%)
Ent. aerogenes	9(1.51%)			*Eubacterium* spp.	21(3.51%)
Flavi oryzihabitan	15(2.51%)			*Fusobacterium* spp.	12 (2.01%)
Klebsiella pneumoniae	12 (2.01)				
Citrobacter spp.	12 (2.01%)				
E.vulneris	102(16.92%)				
Staph. spp.					

Table 4. Comparision of Normal flora of infants & Gastroenteritis patients

	Control			Gastroenteritis Infants		
Bacteria	GPC	GNB	GPB	GPC	GNB	GPB
Percentage	40.30	15.15	43.55	49.25	30.65	20.10

Table 5. Characterisation of diarrhoegenic *E.coli* by serotyping

Types of Strain	Gastroenteritis cases	Control
ETEC	5.8	3.9
EPEC	12.1	5.1

Table 6. Sensitivity in GPC to different group of Antibiotics

Antibiotics	Cephalosporin	Macrolides	Glycopeotides	Aminoglycoside	Penicillin	Quinolone
Percentage	0	0	33.3%	33.3%	33.3%	0

(Table 4). Enteropathogenic *E. coli* are more than that of enterotoxigenic type among the identified *E.coli* from the children suffering from gastroenteritis.

DISCUSSION

The vanous risk factors for gastroententls were broadly classified under different sociodemographic variables i.e age, sex, feeding habits, socioeconomic status etc.

Sex -wise distribution of patients in this study, concluded that percentage of infection was found to be more in males which corroborates with the findings of Ahmed *et al.*[4], Price *et al.*[5]. Among 4 different age group children belonging to age group (>1 yr) suffered significantly more than other age group. Similar results were found by Gupta *et al.*[7] and EI-Gilany *et al.*[8] Considering the socioeconomic status of the patients from LIG and the patients who depend on the well water were affected more which was in agreement with the findings of Gupta *et al.*[7] and Desenclos *et al.*

Feeding habitat of children revealed that commercial milk fed babies were more prone to Diarrhea. Similar results were reported by Donovan SM[10] and Mackie *et al*.[11]

Table 2 depicts the occurrence of bacterial and viral isolates obtained fr.om stool culture. Only bacterial agent acconts more (70%) than the viral agent (bacteria+virus) (30%) which corroborates with the finding of Loening *et al*[12] and Haffejje *et al.*[13] Among the viral agents detected, the rota virus infection was more (23.33%) than adeno virus (6.67%). The present finding was also supported by previous workers Orlandi *et al.*[14] and Filho *et al.*[15] From the *E. coli* identified from the diarrheal cases (12.1 %) enteropathogenic sreogroup of *E. coli* (EPEC) and 5.8% were infected with enterotoxigenic *E.coli* (ETEC).This finding corroborates with the findin~s of Robins *et aL.* 19 and Koomhoof *et al*[2].

The antibiogram study of GPC to different antibiotics (Table 6) revealed that the sensitivity of GPC to glycopeptides (vancomycin). aminoglycosides (amikacin, gentamicin) and penicillin (ampicillin, ampicillin! sulbactum) were (33.3%), (33.3%), (33.3%) respectively. The finding was also supported by Domjanovic *et al.*[21] and Hossam AL *et al.*[22] The antibiogram study of GNB to different antibiotics (table-7) showed that GNB were mostly sensitive (47%) to carbapenems (imipenems) and mostly all the bacteria were sensitive (23%) and (19%) to third generation. of cephalosporin (cephotaxime, ceftazidime, cephoperazone) and quinolones (ciprofloxacin, gatifloxacin, ofloxacin, norfloxacin) respectively. This result was also supported by Cabera *et al.*[23] that third generation of cephalosporin were most effective In treatment of enteric diseases.

ACKNOWLEDGMENTS

This work was supported by Dr. P.Panigrahi, Associate Professor of Pediatrics and Epidemiology and Preventive Medicine, University of Maryland, school of Medicine, USA, and Dr L. Pradhan, Pediatrician, Capital Hospital, Bhubaneswar, Orissa.

REFERENCES

1. Sheath Mini Reeta. Comp leme"ntary associated diarrhea. *Indian J. of Paediatr,* 2006; **73**(1): 61-64.
2. Saravanan P, Ananthan S, Ananthasubra manian M. Rota Virus infection among infants and young children in Chennai,south India, *Indian J of medicaL Microbiol,* 2004; **22**(4) : 212-221.
3. Current Index of Medical specialist, 2004; **86**: 15.
4. Ahmed M., Billo AG. , Murtaza G., Risk factors of persistent diarrhea in children below five years of age. *J. Pak Med Assoc,* 1995; **45**: 290-292.
5. Price V.J. Diagnosis and management of diarrheal disease in Dhaka. J. *of Royale Naval Medical Service* ,1947; **83**(3) :121-126.
6. Bauer Aw, Kirby WMM,Sherris JC,Turck M. Antibiotics susceptibility testing by a standardized single disc method. *Amer J Clinical Pathol* ;**45**: 493-501.
7. Gupta P, Murali M.V, Seth A . Epidemiology of diarrhea in Urban Slum. *Ind. Ped.*, 1998 ; **35**(2): 147-151.
8. EI-Gilany, AH, Hammad, S. Epidemiology of diarrheal diseases among children under 5 years in Dakahlia. *East Mediterr. Health J,* 2005; **11**(4): 762-75
9. Desenclos Jc., Zergabachew A, Desmoulin, B., Chouteau L. Desve. Gand Admassu M .. Clinical, Microbiological and antibiotic

susceptibility patterns of diarrhea in Ethiopia. *J Trap. Med Hyg.*, 1998; **91**(6) : 296-301.

10. Donovan SM.,Oddle J. Growth factors in milk as mediators of infant development. *Annu Rev Nutr.*, 1994; **14**: 147-67.
11. Mackie Roderick, Saghir, Abdelghani and H Rex Gaskins.Developmental microbial ecology of the neonatal gastrointestinal tract. *American J of Clinical Nutrition.* 1999; **69**: 1035-1045.
12. Loening WE, Coovadia, YM, Vandenende J. Etilogical factors of infantile diarrhea. *Ann Trap pediatric,* 1989; **9**(4): 248-55.
13. Haffejee 1.E , Moosa, A Rota virus in Indian (Asian) South African infants with acute Gastroenteritis. *Ann Trap Paediatric.,* (1990)*;* **10**: 165-72.
14. Orlandi P. , Magalhal,Lf-G.F , Matos,N.B. Penatti, M. Nogueira, P.A ,Silva L.H.P.D.A Etiology of diarrheal infection in child in Western Amazon region, Brazil. *Brazilian J of medical and biological research.,* 2006; 39: 507-517.
15. Filho E.P. Da costa faria N.R. Fialho, AM. De Assis, Rs. Almedia M.M., Rocha, M., Galvao, M. Dos Santos, F.S., Barreto, M.L., Laite, J.P. Adenovirus associated with acute gastroenteritis in hospitalized and community children up to 5 years in Brazil. *J Med Microbial.,* 2007; **56**(3): 313-9.
16. Khanduja, P.C., Bhargav, K.C., Gour, H.K., Mallik, D.M .. Etiological aspect of diarrhea in infants ad children under 5 years. *Ind. J Pediat.,* 1969; **36**: 237.
17. Bitterman, R., Rosen, P. ,Barkin, RM. Acute gastroenteritis and Constipation .Concepts and clinical practice. *3rd Ed* 1992; 1537.
18. Vungugen T., VAN Pungle , Chinhle Hug, Weentra VB. ,Khan H NG olyengia. Detection and characterization of diarrheagenic E coli from young children, in Hanoi, Vietnam. *J of clinical Microbial.,* 2005; **43:** 755-760.
19. Robins-BroweneRM. Stillcs Miliotis MD, Richardson NJ., Koornhoof H.J.Freiman 1. Schoub Bd , Lecatsaa, G., Hartman, E. Summer diarrhea in African infants and children *Archives of Disease in child,* 1980; **55**: 923-928.
20. Koomhof Hl Robins-Browne RM., Richardson, N.J. , Cassel R., Etiology of infantile enteritis in South Africa. *Isr. J Med Science.*, 1979; **15**: 341-347.
21. Damjanovic V, Furtado M, Patmore M. Antibiotic sensitivity of enteropathogenic bcteria isolated from patients in Sarjarh Hospital.. *J Hyg (LOND.),* 1984 ;**92**:205-8.
22. Hossam AI- Tatrari, Nahed Abdel-Haq, Pimpanada chearskul, Basim asarnr. Antibiotic for treatment of resistant Gram- positive coccal infection. *Indian J Pediatr*, 2006; **73**(4): 323-334.