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Report on
"A Perspective Study on the Etiology of Diarrhoea in Kathmandu Valley"

Sukraraj Tropical
&
Infectious Disease Hospital
Teku, Kathmandu

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Report on

"A Perspective Study on the Etiology of Diarrhoea in Kathmandu Valley"

Investigators:

Principal investigator: Dr. Basu Dev Pandey

Co-investigators: Dr. L. B. Thapa

Dr. Achyut Bhattarai

Dr. Nirmal Rimal

Address: Sukraraj Tropical and Infectious Disease Hospital (STIDH)

Teku, Kathmandu, Nepal

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"A Perspective Study on the Etiology of Diarrhoea in Kathmandu Valley"

Abstract

One hundred and eighty-one patients attended the gastroenteritis ward of Sukra Raj Tropical and Infectious Disease Hospital (STIDH) with acute diarrhoea and were investigated for etiology of diarrhoea. Bacterial pathogens were isolated among 33% of the patients. Among them enteropathogenic *Escherichia coli* (E.coli) was isolated in 8.28%, *Shigella* species in 13.25% and *Vibrio cholerae* in 1.1% of the patients. Mixed infections with bacterial pathogens, helminthes and protozoan parasites were commonly observed in the study. *Trichuris trichiuria* was detected in 27.6%, hookworm in 12.7% and *Ascaris lumbricoides* in 11.04%. *Entamoeba histolytica* and *Giardia lamblia* each in 11.7 and 7.73 of the patients, respectively. A large number of *Cryptosporidium* (7.73%) and *Cyclospora* species (3.86%) commonly present in immunocompromised patients, were also detected in acute diarrhoeal cases. The results showed that widest ranges of bacterial pathogens were isolated from the inhabitants of Kathmandu prior to monsoon. These findings indicate that the bacterial pathogens especially different species of. *E. coli*, *Shigella* and protozoan parasites need to be given additional attention in the diagnosis and treatment of acute diarrhoea.

Key words: Diarrhoea, Bacterials pathogens, Protozoa, Kathmandu

Introduction

Nepal has one of the lowest standards of health status in South Asia. Diarrhoea is the most common illness among children causing highest numbers of mortality and morbidity in the developing world (2). The highest risk of diarrhoea occurs in the first 5 years of life (2). However, it is also a serious public health problem among adults causing significant levels of morbidity during early summer and the rainy season in Nepal (7). The incidence of diarrhoeal diseases in Nepal tends to rise sharply during warm summer months with the peak in July/August. However, increased incidence of food borne gastroenteritis does occur each year in April/May, followed by waterborne gastroenteritis and cholera in the beginning of rainy season (7). Ministry of Health, Nepal has given top priority for the control and prevention of diarrhoeal diseases. This study will provide base line information and guidelines for the policy-making intervention, and preparation of management protocol for diarrhoeal diseases.

Sukraraj Tropical and Infectious Disease Hospital (STIDH) is the only infectious disease hospital attending diarrhoea patients among the adult population located in Kathmandu, Nepal. A total number of 4497 patients with diarrhoea were admitted in the year of 2000 in STIDH. Although several studies have been conducted in the past, there are no clear figures concerning etiology of diarrhoea in the adult population (4,6,7). At present, there is no well-established protocol for the management of diarrhoeal diseases. In this situation, there is an urgent need to carry out etiological study on diarrhoeal disease to provide base-line information on the causes of diarrhoeal disease and prepare of national protocol for the management of diarrhoea.

The present study is one of the first studies to assess diarrhoea in the adult population in Nepal involved 181 patients with acute diarrhoea who attended STIDH in April-May 2001. The primary aim of this study was to determine the etiology of diarrhoea among the inhabitants of Kathmandu Valley.

Objective of the Study

General : The general objectives of this study is to find out the causes of diarrhoeas among the inhabitants of Kathmandu.

Specific : The specific objectives of this study:

- To find out the etiology of diarrhoeas
- To find out the age and sex wise distribution of diarrhoeas
- To find out the seasonal distribution of diarrhoeas
- To know the average length of hospital stay of diarrhoea patients
- To know the locality wise distribution of diarrhoea patients
- To find out the episode of diarrhoea per year
- To recommend possible interventions, referrals and other programme for future study

Project Design and Methodology

The study was conducted according to the following procedures:

a. One hundred and eighty one patients was included in the study.
b. Inform consent was taken.
c. Demographic profile of the patient recorded using the prepared questionnaire.
d. Patient was examined and degree of dehydration assessed.
e. Stool and blood samples was collected.
f. Samples was examined for parasitic infestation, protozoa, bacterial, fungal and viral causes by microscopic, culture and serological method.
g. Appropriate treatment was given to the patient.
h. Health education was given for the prevention of diarrhoeal diseases to the family members and the patients.
i. Results was tabulated, analyzed, recommendation was prepared.

Methodology in details

Patients admitted to the STIDH with diarrhoea excluding systemic illness were enrolled in the study. Set questionnaires were completed by the staff of STIDH after obtaining written consent from the patients as mentioned earlier. Demographic profile was recorded and degree of dehydration was assessed. Stool samples were collected and based on the degree of dehydration appropriate treatment was started to the patients. Samples were processed for routine parasitological, bacteriological and culture examination. Treatment was reviewed after receiving the laboratory reports on the stool.

Parasitological test

The stool samples were collected in the morning from 181 older than 16 years patients in STIDH and were examined within 2- 4 hours of collection. Direct smears were

prepared with normal saline, iodine and sucrose solution. Two and half percent of potassium dichromate solution and modified Ziehl Nielsen staining were applied to differentiate other protozoan parasites mainly *Cryptosporidium, Isospora* and *Cyclospora* (3). Formal ether concentration method was also used to identify other intestinal parasites.

Bacteriological test

Samples received in laboratory were cultured immediately to identify pathogenic microorganism such as *Vibrio cholerae*, *Salmonella*, and *Shigella* and *Escherichia coli* by using bacteriological method described by Mackie and McCartney, 1995 (In Medical Microbiology, Fourteen edition, 1995 Churchill Livingstone Medical Division of Longman Group Ltd). Stool samples were inoculated into Thiosulphate Citrate Bile Salt Sucrose (TCBS) Agar, MacConkey agar, Eosin Methylene blue (EMB) agar as well as Salmonella (SS) agar into enrichment media such as Selenite F, Rappaport and Alkaline peptone water. After 24 hours of incubation, Rappaports and Selenite F broth was subcultured into MacConkey and Salmonella Shigella agar. Alkaline peptone water was subcultured into TCBS agar after 6 hours and incubated overnight at 37^0C. Typical characteristic Vibrio like colonies was seen on TCBS agar were subcultured on nutrient agar and identified by biochemical test. Biotypes of *V. cholerae* were performed using Voges-Proskauer (VP), polymyxin B sensitive and Haemolysin production test. Biochemically confirmed *V. cholerae* was subjected for serotyping. Serotyping was carried out with the use of monoclonal antiserum raised against *V. cholerae* manufactured by Denka Seiken, Japan as per manufacturer protocol.

Both lactose and non-lactose fermenting colonies from MacConkey and SS Agar were subjected to detailed study. Biochemical and serological tests were used to differentiate pathogenic *E. coli*, *Salmonella* and *Shigella* species. All culture growth microorganism were also confirmed by using Oapi 20E an *invitro* diagnostic biochemical kit manufactured by bioMerieux sa 69280 Marcy/ Etoile-France.

Statistical Methods

Differences between values were examined using students t-test and chi-square test analysed using SPSS version 10, Stastistical Analysis Softwere, 2000.

Results

A total of 181 patients aged 16 years and above attending the STIDH between April-May with acute diarrhoea was included in this study (Table-1). The patient were attended with an average of 10-15 loose motions before coming to hospital. Out of them 60% of the cases were female (P = < .0001) and the highest numbers (40 %) were in the 20-30 age group for both males and females (Fig.1). The main symptoms associated with diarrhoea was combination of abdominal pain (70%), vomiting (62%), and fever (15%) with moderate to severe dehydration (Fig.2). Patients with systemic illness or those that had taken antibiotics prior coming to the hospital were excluded from the study. Since the etiology of diarrhoea has not been studied in adult population, the prevalence of all the enteropathogens including *Cryptosporidium* and *Cyclospora* was also determined.

The patient attended in the hospital reported of having median of 11 hours of loose motion before coming to hospital. The result in Table-2 show that bacterial pathogens were isolated from 33% of the patients. Among them enteropathogenic *E. coli* (EPEC) were isolated from 8.28% (P = .000), *Shigella* species from 13.25% (*Shigella dysenteriae* in 5%, *Shigella flexneri* in 4.41% *Shigella boydi* in 2.2% *Shigella sonnie* in 1.65%) and *Vibrio cholerae* was isolated in only 1.1% of the patients seen during the study period. It showed that *EPEC* was the most commonly isolated single bacterial pathogen followed by *Shigella dysenteriae* in 4.41 % cases. Whereas *Salmonella* species were isolated in 4.41% of the cases. (P = .004).

Most of the patients were infected with more than one organism(Fig.4). Protozoal pathogens *E. histolytica*, *G. lamblia*, *Cryptospridium* and *Cyclospora* were detected in 31% of the patients (Table 3). Out of them 11.6% were infected with *E. histolytica*, 3.86% with *Cyclospora* and 7.73% each with *G. lamblia* and *Cryptosporidium*. These results indicated that *Cryptosporidium* and *Cyclospora* species are also important pathogens causing diarrhoea in Kathmandu. It was also found that *T. trichiuria* infection was the predominant helminthic infection (27.6 %) among these patients (P = .000). On the other hand 13 % patients were infected with hookworm and 11.04% with *A. lumbricoides* (Table 4). The symptoms associated with *Blastocystis hominis* are similar to

those associated with *G. lamblia* both of them isolated in 7.73% of the patients. *Hymenolepsis nana* and *Isospera belli* were detected in 1.1 and 0.55 % of the patients, respectively.

Although *V. cholerae* is the major cause of acute watery diarrhoea during the rainy season, it was isolated 1.1% during the study period. On the other hand the rare pathogens like *Chilomastix mesnili*, *Indamoeba butschlii* were also detected in the range of 1% and *Strongyloides* was detected in 1.65 % of the cases. The present result indicates that there is a seasonal variation on the etiology of diarrhoea in Kathmandu Valley which should be confirmed by more extensive study.

Discussion

In Nepal, the incidence of diarrhoeal diseases rises sharply each year during the early warm summer and rainy season with the epidemic peak occurring in July-August each year (7). However, report indicates that incidence of food borne gastroenteritis does occur each April/May, followed by waterborne gastroenteritis and cholera with the beginning of rainy season (7). This is one of the first studies to assess the etiology of diarrhoea during warm early monsoon among the adult population in Nepal. This study confirms the importance of *E. coli* species, *Shigella*, *E. histoytica*, *G. lamblia* and *Cryptosporidium* as the causing agent of acute diarrhoea in Nepal. Charles W.H. reported similar observation among under 5 populations (4). The low prevalence of cholera in this study is most likely due to season of study. It is important that an etiological study should be continued longitudinally irrespective of age of the patients.

Diarrhoea is usually classified as an inflammatory and noninflammatory since such a classification has therapeutic significance. The other species of *E.coli*, which was not isolated in this study like enteroaggregative E. coli, may be an important cause of acute diarrhoea in developing countries (1). More extensive study is required to rule out such a possibility as a cause of diarrhoea. Inflammatory diarrhoeas that may be associated with higher mortality can be studied by stool culture. Several inflammatory marker like faecal leucocytes, occult blood, and fecal lactoferrin could be used as a predictor of invasive pathogen of enteric inflammation (5).

The source of drinking water, knowledge and attitude of the patients and the family members were assessed in this study. As shown in Fig. 3, 80% of the patients reported to use their tap water as a source of drinking water, which is highly polluted with high coliform and low chlorine concentration in Nepal (7). Only 23% families boil water for drinking which could be one of the major predisposing factors for diarrhoea. The 45% of the patients still believe that the diarrhoeal diseases are not communicable. Contaminated public water supplies probably contributed significantly to sustained transmission of disease in Kathmandu.

Recommendation

Diarrhoea is still the most common illness among children causing highest numbers of mortality and morbidity in the developing world like Nepal. The highest risk of diarrhoea occurs in the first 5 years of life. However, it is also a serious public health problem among adults causing significant levels of morbidity during early summer and the rainy season in Nepal. The present study showed that diarrhoea is also a important problem in the adult population in Kathmandu Valley. The information obtained from this study showed that the source of polluted drinking water and lack of knowledge of the patients and the family members on the etiology of diarrhoea are the major factors for increased cases of diarrhoea. Health education regarding the etiology of diarrhoea and its prevention could be important steps in preventing diarrhoeal disease. Eighty percent of the patients reported to use tap water as a source of drinking water, which is highly polluted with high coliform and low chlorine concentration in Nepal. Fourty five percent of the patients still believe that the diarrhoeal diseases are not communicable. Contaminated public water supplies probably contributed significantly to sustained transmission of disease in Kathmandu. Only 23% families boil water for drinking which could be other important predisposing factor for diarrhoea. Safe water supply could prevents episodes of diarrhoea, decreased the number of cases and mortality. The findings also indicates that the bacterial pathogens especially different species of. *E. coli*, *Shigella* and protozoan parasites need to be given additional attention in the diagnosis and treatment of acute diarrhoea. The study showed that there are very few people using ORS before coming to the hospitals. Health education and use of ORS is still play vital role for the prevention of dehydration. Most of the patients receiving antibiotics before coming to the hospital. It make major challenge to find the causative agents and to give appropriate treatment. Existing protocol for the treatment of diarrhoea did not match the finding of the patients. Refresher perioding training should be given to the health workers and drug distributors about the management of diarrhoea. Revised national protocol should be available for the management of diarrhoea. Long term monitoring of the situation and continuos study on diarrhoeal disease is important for the control and prevention of diarrhoeal disease in Kathmandu, Valley.

Acknowledgments

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Table. 1 Enteric pathogens identified among 181 patients of diarrhoea from April and May, 2001 in STIDH, Teku, Kathmandu

Serial Number	Enteropathogens	Patients (n=181)	Percentage
1	Enteropathogenic *Escherichia coli*	15	8.28
2	Enterotoxigenic *Escherichia coli*	5	2.76
3	Enteroinvasive *Escherichia coli*	4	2.20
4	Enterohaemorrhagic *Escherichia coli*	2	1.10
5	*Shigella dysenteriae*	9	5.0
6	*Shigella boydi*	4	2.2
7	*Shigella flexneri*	8	4.41
8	*Shigella sonnei*	3	1.65
9	*Salmonella species*	8	4.41
10	*Vibrio cholerae (Ogawa)*	2	1.1
11	*Entamoeba histolytica/dispe*	23	12.7
12	*Cyclospera species*	7	3.86
13	*Cryptosporidium*	14	7.73
14	*Blastocystis hominis*	14	7.73
15	*Ascaris lumbricoides*	20	11.04
16	*Giardia lamblia*	14	7.73
17	*Trichuris trichiuria*	50	27.6
18	Hookworm	23	12.7
29	*Hymenolepsis nana*	2	1.1
20	*Chilomastix mesnili*	3	1.65
21	*Escherichia coli*	12	6.62
22	*Dientamoeba fragilis*	1	5.52
23	*Trichomonas*	1	0.55
24	*Camphylobacter jejuni*	1	0.55
25	*Strongyloides*	3	1.65
25	*Isospora belli*	1	0.55
26	*Iodamoeba butschlii*	3	1.65

Tabel 2: Bacterials Isolates

Escherchia coli	**26**	**14.36**
EPEC	15	
ETEC	5	
EIEC	4	
EHEC	2	
Shigella spp.	**24**	**13.25**
Shigella dysenteriae	9	
Shigella boydi	4	
Shigella flexneri	8	
Shigella sonnei	3	
Salmonella spp.	**8**	**4.4**
Salmonella paratypi B	3	
Salmonella typhi	2	
Salmonella typhimurium	2	
Salmonella barielly	1	
Vibrio cholerae (Ogawa)	**2**	**1.1**
Camphylobacter jejuni	**1**	**0.55**

Table 3: Protozoans

Amoebas		
Entamoeba histolytica/dispar	23	12.7
Blastocystis hominis	14	7.73
Entamoeba coli	12	6.62
Iodamoeba butschlii	3	1.65
Flagellates		
Giardia lamblia	14	7.73
Dientamoeba fragilis	1	0.55
Chilomastix mesnili	3	1.65
Trichomonas hominis	1	0.55
Coccidians		
Cryptosporidium parvum	14	7.73
Cyclospera spp	7	3.86
Isospora belli	1	0.55

Table 4: Helminths

Nematodes		
Trichuris trichiuria	50	27.6
Hookworm	23	12.7
Ascaris lumbricoides	20	11.04
Strongyloides	3	1.65
Cestodes		
Hymenolepsis nana	2	1.1

Fig. 1

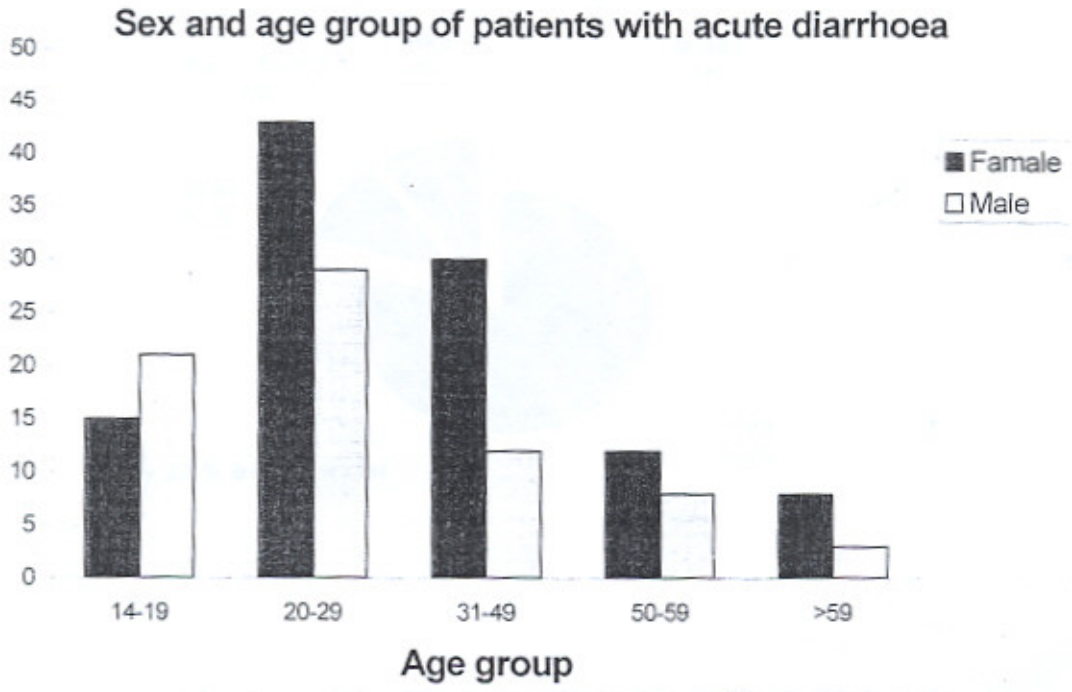

Sex and age group of patients with acute diarrhoea
50
45
40
35
30
25
20
15
10
5
0
Famale
Male
14-19
20-29
31-49
50-59
>59
Age group

Fig. 2

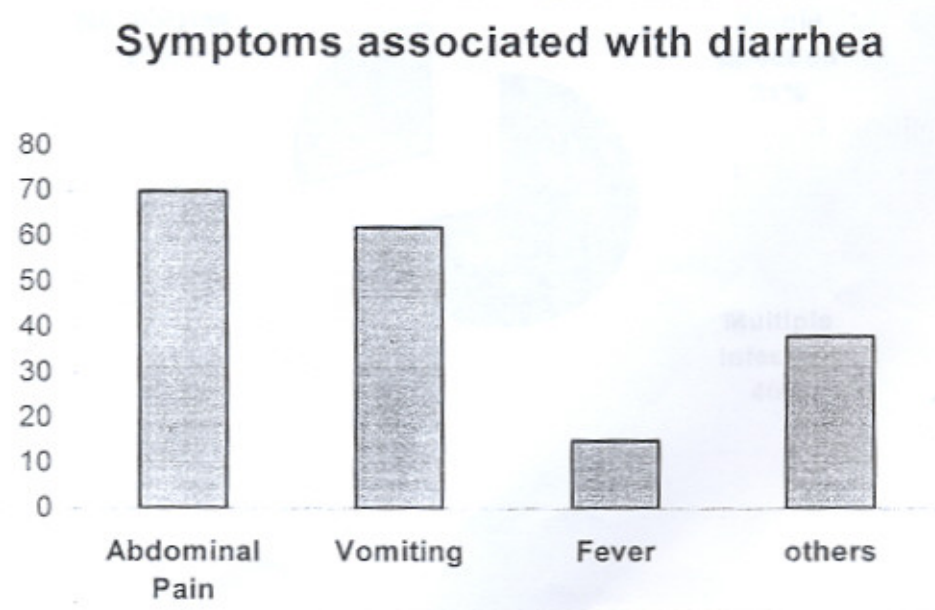

Symptoms associated with diarrhea
80
70
60
50
40
30
20
10
0
Abdominal Pain
Vomiting
Fever
others

Fig. 3

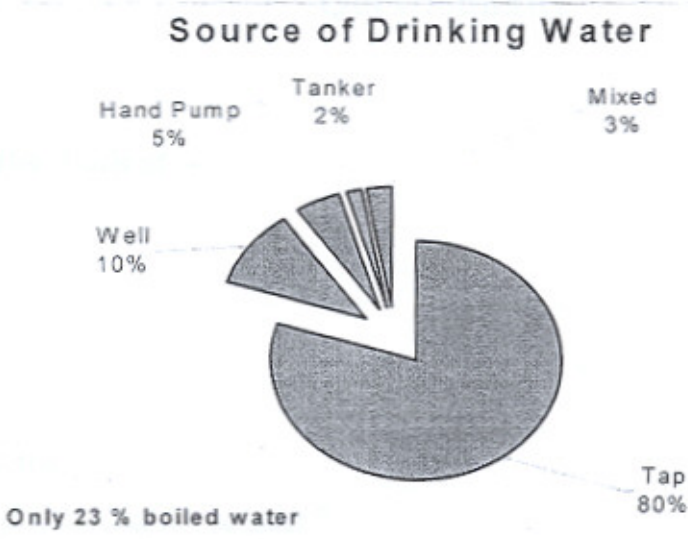
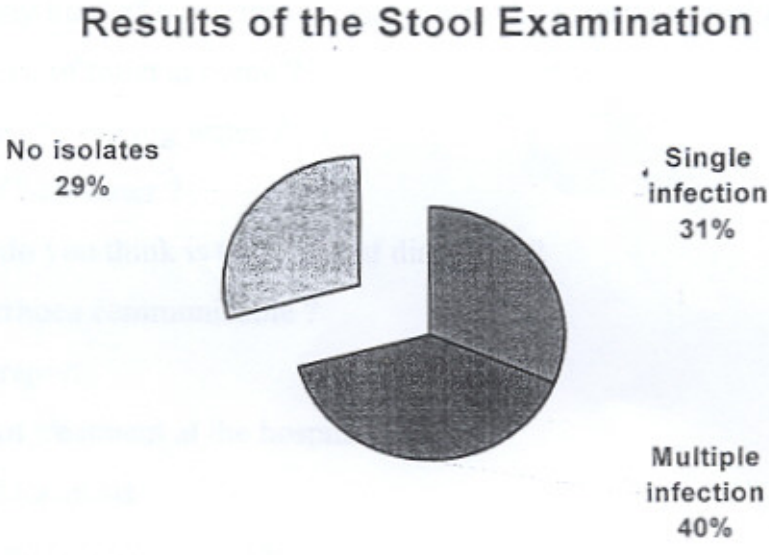

Source of Drinking Water
Hand Pump 5%
Tanker 2%
Mixed 3%
Well 10%
Tap 80%
Only 23 % boiled water

Fig.4


Results of the Stool Examination
No isolates 29%
Single infection 31%
Multiple infection 40%

Questionnaire

Name of the Patient :-

1. Sex
2. Age
3. Address
4. Symptoms
5. When did the diarrhoea start ?
6. No. of loose stools/ day
7. Type of diarrhoea
8. Severity of dehydration
9. How many episode of diarrhoea did you have/ year ?
10. Are there other members in the family suffering from diarrhoea ?
11. Did the patient receive any 'Jevan Jal' prior to coming to the hospital ?
12. Was any kind of medicines given prior to the patients arrival at the hospital?
13. Presence of toilet at home ?
14. Source of drinking water ?
15. Use of boil water ?
16. What do you think is the cause of diarrhoea ?
17. Is diarrhoea communicable ?
18. Stool report
19. Type of treatment at the hospital
20. Final Diagnosis
21. Date and time of discharge
22 Total duration of hospital study (in hours)