Infection and Drug Resistance

Dovepress
open access to scientific and medical research

Open Access Full Text Article

ORIGINAL RESEARCH

Clinical features, risk factors, and impact of antibiotic treatment of diarrhea caused by *Shigella* in children less than 5 years in Manhiça District, rural Mozambique

This article was published in the following Dove Press journal:
Infection and Drug Resistance

Infection and Drug Resistance downloaded from https://www.dovepress.com/
For personal use only.

Delfino Vubil[1]
Sozinho Acácio[1,2]
Llorenç Quintò[3]
Clara Ballesté-Delpierre[3]
Tacilta Nhampossa[1,2]
Karen Kotloff[4]
Myron M Levine[4]
Pedro Alonso[1]
James P Nataro[5]
Tamer H Farag[4]
Jordi Vila[3,6]
Inacio Mandomando[1,2]

[1]Centro de Investigação em Saúde de Manhiça (CISM), Maputo, Mozambique; [2]Instituto Nacional de Saúde (INS), Maputo, Mozambique; [3]ISGlobal, Hospital Clínic - Universitat de Barcelona, Barcelona, Spain; [4]Center for Vaccine Development (CVD), University of Maryland, School of Medicine, Baltimore, MD, USA; [5]Department of Pediatrics, University of Virginia School of Medicine, Charlottesville, VA, USA; [6]Department of Clinical Microbiology, Centre for Biomedical Diagnosis, Hospital Clinic, Barcelona, Spain

Correspondence: Inacio Mandomando
Centro de Investigação em Saúde de Manhiça (CISM), Rua 12, Bairro Cambeve, P.O. Box 19292, Maputo, Mozambique
Tel +258 21 810 002
Fax +258 21 810 181
Email inacio.mandomando@manhica.net

Objectives: During the period from December 2007 to November 2012, the epidemiology of diarrhea caused by *Shigella* was studied among children <5 years of age residing in Manhiça District, Southern Mozambique.

Materials and methods: Children from 0 to 5 years with moderate-to-severe diarrhea (MSD) and less severe diarrhea (LSD) were enrolled along with matched controls (by age, gender, and neighborhood). Age-stratified logistic regression analyses were conducted to identify clinical features and risk factors associated with *Shigella* positivity in cases of diarrhea. The impact of antibiotic treatment was assessed for patients with known outcome.

Results: A total of 916 cases of MSD and 1979 matched controls, and 431 cases of LSD with equal number of controls were enrolled. *Shigella* was identified as significant pathogen in both cases of MSD and LSD compared to their respective controls. *Shigella* was detected in 3.9% (17/431) of LSD compared to 0.5% (2/431) in controls (P=0.001) and in 6.1% (56/916) of MSD cases compared to 0.2% (4/1979) in controls (P<0.0001), with an attributable fraction of 8.55% (95% CI: 7.86–9.24) among children aged 12–23 months. Clinical symptoms associated to *Shigella* among MSD cases included dysentery, fever, and rectal prolapse. Water availability, giving stored water to child, washing hands before preparing baby's food, and mother as caretaker were the protective factors against acquiring diarrhea caused by *Shigella*. Antibiotic treatment on admission was associated with a positive children outcome.

Conclusion: *Shigella* remains a common pathogen associated with childhood diarrhea in Mozambique, with dysentery being a significant clinical feature of shigellosis. Adherence to the basic hygiene rules and the use of antibiotic treatment could contribute to the prevention of most of diarrhea due to *Shigella*.

Keywords: *Shigella*, moderate-to-severe diarrhea, less severe diarrhea, epidemiology

Introduction

Diarrhea is the leading cause of mortality in children <5 years with its long-term impact on growth and cognitive development.[1] Although the burden is greater in low-income populations, acute infectious diarrhea is also a common cause of outpatient visits and hospital admissions in high-income regions and is an important health problem globally.[2] Associated risk factors for infection include poor living standards, overcrowding, inadequate sanitation, and poor hand hygiene. These factors may result in a significant disease burden and economic effect due to direct medical costs, loss of work, lower quality of life, and mortality.[3]

submit your manuscript | www.dovepress.com
Dovepress
http://dx.doi.org/10.2147/IDR.S177579

© 2018 Vubil et al. This work is published and licensed by Dove Medical Press Limited. The full terms of this license are available at https://www.dovepress.com/terms.php and incorporate the Creative Commons Attribution – Non Commercial (unported, v3.0) License (http://creativecommons.org/licenses/by-nc/3.0/). By accessing the work you hereby accept the Terms. Non-commercial uses of the work are permitted without any further permission from Dove Medical Press Limited, provided the work is properly attributed. For permission for commercial use of this work, please see paragraphs 4.2 and 5 of our Terms (https://www.dovepress.com/terms.php).

The high burden of the disease is also associated with the wide range of recognized enteric pathogens such as virus, bacteria, and parasites that may cause diarrhea.[4] Among them, *Shigella* infections remain a global public health concern, in resource-limited countries where the disease may cause as many as 163 million episodes of diarrhea and over a million deaths annually with the majority (60%) occurring in children under 5.[5] Due to the high disease burden and endemicity of *Shigella* in resource-limited countries, the WHO has set the development of candidate vaccines against *Shigella* as priority.[6] Thus, considering the efficacy of *Shigella* vaccine, it is necessary to have reliable estimates of the burden and epidemiology of disease in targeted endemic areas, including age-specific incidence data and risk factors associated with *Shigella* infections.

Primarily, shigellosis is transmitted from person to person through the fecal–oral route, but it may also spread indirectly by fecal contamination of water or food.[7] Signs/symptoms of individuals who are infected with *Shigella* expand fever, painful bloody diarrhea, and stomach cramps.[8] WHO recommends that all episodes of diarrhea with blood in the stool must be treated with antibiotics.[9] Antimicrobial therapies reduce the period and intensity of disease symptoms, decrease excretion of bacteria, and prevent potentially lethal complications. Decreasing the bacterial load excreted by a child with dysentery also reduces the probability of fecal–oral transmission to close contacts, such as neighbors, friends, or members of the child's household.[10]

In Mozambique, epidemiologic data on diarrhea caused by *Shigella* remain scarce and existing data are limited to few studies.[11,12] Moreover, the contribution of *Shigella* to less severe diarrhea (LSD), the risk factors, and the impact of antibiotic treatment on patient outcome remain unknown. We hereby report the epidemiology of diarrhea due to *Shigella* among children <5 years of age enrolled between 2007 and 2012 in Manhiça district as part of the Global Enteric Multicenter Study.[13]

Materials and methods

Study area and population

The study was conducted by the Manhiça Health Research Centre (Centro de Investigação em Saúde de Manhiça – CISM) in Manhiça district, a rural area of Maputo province in southern Mozambique. The climate is subtropical with warm and rainy season from October to April and a fresh and dry season for the rest of the year. A round-the-clock morbidity surveillance system, covering pediatric inpatient and outpatient visits, was established in 1998 in the Manhiça District Hospital in a joint collaboration with CISM. Clinical data are routinely collected from all children under 15 years seeking health care. HIV infection is among the highest worldwide, with prevalence rates in women in childbearing age as high as 40% in the district.[14] Diarrhea is the third leading cause of hospital admission among children aged 0–14 years and the fourth cause of death in children from 12 to 59 months.[15] Additionally, since 1996, CISM has been running a Health Demographic Surveillance System (HDSS) for vital events and migrations in the population living within the study area covering approximately 95,000 inhabitants during the study period. In 2014, the study area was expanded to the whole district, and currently, 183,000 inhabitants are under DSS follow-up. Each person living within the DSS study area is issued a unique Permanent Identification Number that describes the geographic location, household number, and personal number within the household. A full description of the geographic and sociodemographic characteristics of the study community has been detailed elsewhere.[16]

Study design

The present analysis is part of a prospective case–control study of diarrhea conducted in Manhiça district from December 2007 to November 2012 to estimate the burden and etiology of moderate-to-severe diarrhea (MSD) in children <5 years of age to guide future interventions. To assess the role of *Shigella* in LSD, cases of LSD were enrolled from November 3rd 2011 to November 2nd 2012. Age in months was stratified in three strata (0–11, 12–23, and 24–59). Clinical and epidemiologic data were collected over the study period. Cases of diarrhea included in the study corresponded to children <5 years of age living within the HDSS area presenting to Manhiça district hospital with symptoms of diarrhea, defined as three or more loose stools in the last 24 hours. Clinicians assessed each child with diarrhea for eligibility; to be included, the episode had to be new (onset after ≥7 diarrhea-free days), acute (onset within the previous 7 days), and fulfill at least one of the following criteria for MSD: sunken eyes (confirmed by parent or caretaker as more than normal); loss of skin turgor (abdominal skin pinch with slow [≤2 seconds] or very slow [≥2 seconds] recoil); intravenous hydration administered or prescribed; dysentery (visible blood in loose stools); or admission to hospital with diarrhea or dysentery.[17] For each child with diarrhea, one to three healthy control children (no story of diarrhea in the previous 7 days, matched by age, sex, and neighborhood) were randomly selected using the HDSS database and enrolled within 14 days of presentation of the corresponding index case.

Sample collection and laboratory analysis

Fecal samples from cases were collected within 12 hours of registration of the diarrheal episode, and control samples within 14 days after case enrolment. Once collected, samples were kept in a cool box until processed. Each fecal specimen comprised a whole stool specimen (in screw top fecal specimen cups carried in Styrofoam boxes with cold packs), a fecal swab in Modified Cary Blair medium in a plastic screw top test tube, and a fecal swab in buffered glycerol saline in a screw top test tube.[13] Additionally, if antibiotics were to be given to patients before the production of stool sample, two rectal swabs were obtained for bacterial culture pending passage of the whole stool for the remaining assays. Stool samples were tested for the presence of bacterial, viral, and parasitic pathogens by culture, ELISA, and PCR as appropriate.[18] In the case of S*higella,* species were identified by colonies morphology upon culturing in MacConkey and XLD followed by slide agglutination with specific antisera (Denka Seiken Co., Ltd., Tokyo, Japan).

Data analysis

The software for analysis was Stata/SE version 14.1 (STATA Corporation, College Station, TX, USA) and the package coxphf from R version 3.2.2. All analyses were stratified by age. The frequency of *Shigella* isolation was calculated dividing the number of *Shigella* positivity by the total number of children enrolled over the study period with known culture result for *Shigella*. The pathogenicity of *Shigella* was assessed comparing the isolation rate between cases of diarrhea and control group. Logistic regression models were used to evaluate associations. All models were estimated with the penalized likelihood according to the Firth's approach.[19,20] Multivariable models were estimated by forward-stepwise selection from covariates with a *P*-value<0.20 in the crude models and no more than 5% of missing values. Significant levels for removal and addition in the multivariable models were 0.10 and 0.05, respectively, by Wald test. The association between MSD and *Shigella* was assessed at crude level for all pathogens and for *Shigella* adjusted for other significant pathogens and pairwise interactions between each one of them with *Shigella*. Analysis of sign/symptoms and sociodemographic factors associated with *Shigella*-MSD vs other types of MSD was performed among MSD cases. Interactions between significant covariates in the multivariable model were also assessed.

Ethical approval

The study is part of the Global Enteric Multi-Center Study (GEMS), which was approved by the Institutional Review Board at the University of Maryland School of Medicine, USA and the National Bioethics Committee for Health of Mozambique and Hospital Clinic, University of Barcelona, Spain. A written informed consent was obtained from children's mothers or caretakers for participation in the study.

Results

Shigella and association with diarrhea

During the study period, a total of 916 cases and 1979 controls for MSD (December 9, 2007 to November 2, 2012), and 431 LSD cases and their respective 431 matched controls (November 3, 2011 to November 2, 2012) were enrolled. S*higella* was isolated in 6.1% (56/916) of MSD cases compared to 0.2% (4/1979) in controls (*P*<0.0001) and in 3.9% (17/431) of LSD compared to 0.5% (2/431) in control group (Table 1). In addition, *Shigella* infection was predominantly detected during rainy season (October–March) compared to dry season (Figure 1). The incidence rate of *Shigella*-associated MSD was 0.61 per 100 child-year (95% CI: 0.48–0.74) with an attributable fraction of 8.55 (95% CI: 7.86–9.24) in children aged 12–23 months. A strong association of *Shigella* and MSD was found by both crude and pathogen-adjusted multivariate analysis for all age groups especially in children aged 24–59 months where adjusted odds ratio (aOR) of 190.92 (95% CI: 23.80–24565.84; *P*<0.0001) was observed as shown in Table 2.

Clinical presentation

The clinical characteristics (signs and symptoms) of children infected with *Shigella* were compared with non-MSD *Shigella* using crude and multivariate analysis and presented in Table 3. The odds of having *Shigella* infections were

Table 1 Detection rate of *Shigella* among children 0–59 months of age with diarrhea (MSD and LSD) and their matched controls in Manhiça district, December 2007 to November 2012

Age groups	Case/control		*P*-value
	Controls, n (%)	Cases, n (%)	
MSD			
0–11 months	1/1047 (0.1)	6/495 (1.2)	0.002
12–23 months	2/629 (0.3)	21/276 (7.6)	<0.0001
24–59 months	1/303(0.3)	29/145 (20)	<0.0001
Total	4/1979 (0.2)	56/916 (6.1)	<0.0001
LSD			
0–11 months	0/155 (0)	1/155 (0.7)	0.317
12–23 months	1/175 (0.6)	10/175 (5.7)	0.006
24–59 months	1/101 (1)	6/101 (5.9)	0.054
Total	2/431 (0.5)	17/431 (3.9)	0.001

Abbreviations: LSD, less severe diarrhea; MSD, moderate-to-severe diarrhea.

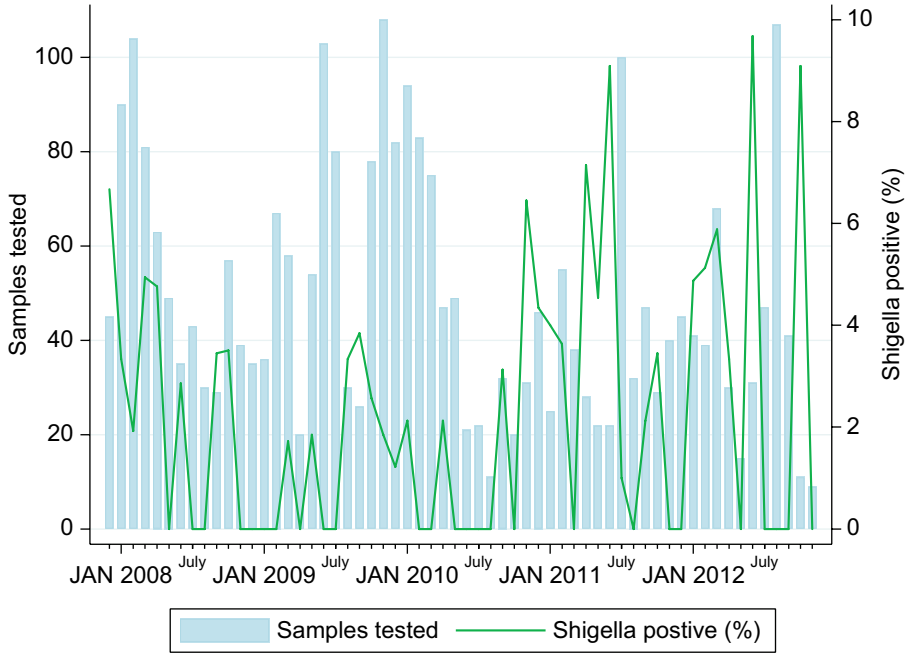

Figure 1 Seasonal distribution of *Shigella* infection in children aged 0–59 months in Manhiça District (December 2007–November 2012).

Table 2 Crude and pathogen-adjusted multivariate analysis of the association of *Shigella* spp. with moderate-to-severe diarrhea among children under 5 years (December 2007–November 2012)

Variables	Group		Crude analysis		Adjusted analysis	
	Controls, n (%)	Cases, n (%)	OR (95% CI)	*P*-value	Adjusted OR (95% CI)	*P*-value
0–11 months						
Shigella spp.	1/1047 (0)	6/495 (1)	22.01 (2.44–2912.20)	0.0028	42 (93.96–5717.57)	0.0008
Cryptosporidium	96/1046 (9)	98/495 (20)	2.89 (2.07–4.06)	<0.0001	3.71 (2.54–5.50)	<0.0001
Giardia	177/1046 (17)	45/495 (9)	0.48 (0.33–0.68)	<0.0001	0.47 (0.31–0.69)	0.0001
Rotavirus	160/1046 (15)	217/495 (44)	5.91 (4.42–8.01)	<0.0001	6.37 (4.69–8.79)	<0.0001
Adenovírus (40/41)	9/1044 (1)	12/493 (2)	3.05 (1.25–7.75)	0.0144	4.43 (1.62–12.57)	0.0038
Nontyphoidal *Salmonella*	6/1047 (1)	6/495 (1)	2.53 (0.81–7.85)	0.108		
Campylobacter	52/1047 (5)	30/495 (6)	1.45 (0.88–2.38)	0.1456		
Sapovírus	29/1047 (3)	9/495 (2)	0.61 (0.26–1.28)	0.1967		
Enteroaggregative *E. coli* (*aaic* only)	59/1047 (6)	40/495 (8)	1.38 (0.89–2.12)	0.1527		
12–23 months						
Shigella spp.	2/629 (0)	21/276 (8)	20.75 (6.61–103.57)	<0.0001	20.6 (6.30–105.59)	<0.0001
Cryptosporidium	60/628 (10)	51/275 (19)	2.63 (1.70–4.08)	<0.0001	2.61 (1.62–4.23)	<0.0001
Giardia	278/628 (44)	74/275 (27)	0.44 (0.32–0.61)	<0.0001	045 (0.32–0.63)	<0.0001
Rotavirus	102/628 (16)	62/275 (23)	1.87 (1.28–2.75)	0.0014	2.07 (1.36–3.16)	0.0007
EAEC (*aatA* only)	30/629 (5)	30/276 (11)	2.14 (1.22–3.80)	0.0079	2.12 (1.15–3.96)	0.0168
Aeromonas spp.	2/629 (0)	3/276 (1)	4.20 (0.82–25.21)	0.0837		
Vibrio cholerae 01	1/629 (0)	4/276 (1)	3.97 (0.69–41.39)	0.1255		
Nontyphoidal *Salmonella*	0/629 (0)	2/276 (1)	15.00 (1.22–2068.84)	0.0336		
Adenovírus (40/41)	12/628 (2)	4/272 (1)	1.08 (0.33–2.97)	0.1416		
24–59 months						
Shigella spp.	1/303 (0)	29/145 (20)	126.56 (17.67–16063.77)	<0.0001	191 (23.80–24565.84)	<0.0001
Giardia	155/303 (51)	50/145 (34)	0.51 (0.32–0.78)	0.0022	0.43 (0.25–0.72)	0.001
EAEC (*aatA*/aaic)	10/303 (3)	1/145 (1)	0.25 (0.03–1.13)	0.0743	0.07 (0.00–0.81)	0.0284
V. cholerae	0/303 (0)	9/145 (6)	23.41 (2.87–3044.31)	0.0008	19.6 (2.48–2523.83)	0.0015
Cryptosporidium	22/303 (7)	11/145 (8)	1.35 (0.60–2.94)	0.4577		
Norovírus	15/303 (5)	5/145 (3)	0.46 (0.16–1.18)	0.1091		
Adenovírus (non-40/41)	8/303 (3)	0/145 (0)	0.11 (0.00–1.01)	0.0519		

Table 3 Clinical presentation of *Shigella*-associated MSD by crude and pathogen-adjusted analysis

Variables	Type of MSD crude analysis				Adjusted analysis	
	Non-*Shigella*-MSD, n (%)	*Shigella*-MSD, n (%)	OR (95% CI)	*P*-value	Adjusted OR (95% CI)	*P*-value
0–11 months of age						
Dysentery	35/489 (7)	1/6 (17)	3.49 (0.56–21.93)	0.1823		
Sunken eyes	279/489 (57)	3/6 (50)	0.75 (0.17–3.35)	0.7096		
Loss of skin turgor	172/489 (35)	2/6 (33)	1.02 (0.22–4.85)	0.9776		
Intravenous rehydration	259/489 (53)	3/6 (50)	0.89 (0.20–3.95)	0.8763		
Hospitalized	344/489 (70)	5/6 (83)	1.55 (0.25–9.52)	0.6369		
Vomiting 3 or more times per day[a]	266/489 (54)	2/6 (33)	0.47 (0.10–2.21)	0.3361		
Very thirsty[a]	345/487 (71)	4/6 (67)	0.74 (0.16–3.53)	0.7079		
Drank much less than usual[a]	70/489 (14)	1/6 (17)	1.62 (0.26–10.05)	0.6027		
Unable to drink[a]	43/489 (9)	0/6 (0)	0.79 (0.04–14.25)	0.8728		
Belly pain[a]	129/489 (26)	2/6 (33)	1.55 (0.33–7.35)	0.5836		
Fever[a]	170/489 (35)	2/6 (33)	1.04 (0.22–4.94)	0.9596		
Irritability or restless attitude[a]	82/488 (17)	3/6 (50)	4.93 (1.10–22.09)	0.0372		
Decreased activity or lethargy[a]	238/488 (49)	1/6 (17)	0.29 (0.05–1.76)	0.1768		
Loss of consciousness[a]	8/489 (2)	0/6 (0)	4.36 (0.23–83.70)	0.3290		
Rectal straining[a]	24/489 (5)	0/6 (0)	1.46 (0.08–26.70)	0.7979		
Rectal prolapse[a]	2/489 (0)	0/5 (0)	17.73 (0.76–413.90)	0.0737		
Cough[a]	331/489 (68)	3/6 (50)	0.48 (0.11–2.13)	0.3329		
Difficulty breathing[a]	54/489 (11)	1/6 (17)	2.18 (0.35–13.55)	0.4034		
Convulsion[a]	13/489 (3)	0/6 (0)	2.72 (0.15–50.69)	0.5036		
Very thirsty[b]	349/487 (72)	4/6 (67)	0.71 (0.15–3.39)	0.6709		
Drinks poorly[b]	60/488 (12)	1/6 (17)	1.93 (0.31–11.99)	0.4796		
Sunken eyes[b]	268/488 (55)	3/6 (50)	0.82 (0.18–3.65)	0.7959		
Wrinkled skin[b]	165/488 (34)	1/6 (17)	0.53 (0.09–3.27)	0.4970		
Irritable or restless[b]	78/489 (16)	2/6 (33)	2.91 (0.61–13.93)	0.1806		
Lethargy or loss of consciousness[b]	238/489 (49)	1/6 (17)	0.29 (0.05–1.76)	0.1782		
Dry mouth[b]	216/488 (44)	1/6 (17)	0.34 (0.06–2.11)	0.2480		
Fast breathing[b]	148/489 (30)	1/6 (17)	0.63 (0.10–3.85)	0.6145		
Undernutrition	48/489 (10)	0/6 (0)	0.70 (0.04–12.62)	0.8092		
Low or very low skin pinch	144/450 (32)	1/5 (20)	0.71 (0.11–4.53)	0.7147		
12–23 months of age						
Dysentery	45/255 (18)	9/21 (43)	3.52 (1.43–8.67)	0.0063	3.99 (1.53–10.39)	0.0046
Sunken eyes	121/255 (47)	6/21 (29)	0.46 (0.18–1.20)	0.1127		
Loss of skin turgor	66/255 (26)	4/21 (19)	0.73 (0.25–2.14)	0.5700		
Intravenous rehydration	113/255 (44)	7/21 (33)	0.65 (0.26–1.62)	0.3553		
Hospitalized	172/255 (67)	12/21 (57)	0.64 (0.26–1.54)	0.3168		
Vomiting 3 or more times per day[a]	124/255 (49)	7/21 (33)	0.55 (0.22–1.36)	0.1953		
Very thirsty[a]	208/255 (82)	15/20 (75)	0.64 (0.23–1.79)	0.3957		
Drank much less than usual[a]	14/255 (5)	1/21 (5)	1.22 (0.21–6.95)	0.8238		
Unable to drink[a]	16/255 (6)	1/21 (5)	1.06 (0.19–6.00)	0.9456		
Belly pain[a]	68/255 (27)	6/21 (29)	1.15 (0.44–2.99)	0.7776		
Fever[a]	84/255 (33)	10/21 (48)	1.85 (0.77–4.45)	0.1676	3.02 (1.15–7.94)	0.025
Irritability or restless attitude[a]	40/255 (16)	1/21 (5)	0.39 (0.07–2.11)	0.2744		
Decreased activity or lethargy[a]	135/255 (53)	9/21 (43)	0.68 (0.28–1.63)	0.3821		
Loss of consciousness[a]	5/255 (2)	0/21 (0)	1.06 (0.06–19.80)	0.9693		
Rectal straining[a]	12/254 (5)	1/21 (5)	1.42 (0.25–8.19)	0.6952		
Rectal prolapse[a]	0/255 (0)	0/21 (0)	–	–		
Cough[a]	159/255 (62)	7/21 (33)	0.31 (0.13–0.78)	0.0130	0.27 (0.10–0.72)	0.0085
Difficulty breathing[a]	21/255 (8)	0/21 (0)	0.25 (0.01–4.33)	0.3435		
Convulsion[a]	12/255 (5)	1/21 (5)	1.43 (0.25–8.22)	0.6918		
Very thirsty[b]	209/255 (82)	15/21 (71)	0.53 (0.20–1.40)	0.1983		
Drinks poorly[b]	15/254 (6)	1/21 (5)	1.13 (0.20–6.41)	0.8897		
Sunken eyes[b]	124/255 (49)	6/21 (29)	0.44 (0.17–1.14)	0.0923		
Wrinkled skin[b]	62/255 (24)	2/21 (10)	0.40 (0.10–1.53)	0.1788		

(*Continued*)

Table 3 (*Continued*)

Variables	Type of MSD crude analysis				Adjusted analysis	
	Non-*Shigella*-MSD, n (%)	*Shigella*-MSD, n (%)	OR (95% CI)	*P*-value	Adjusted OR (95% CI)	*P*-value
Irritable or restless[b]	41/255 (16)	2/21 (10)	0.66 (0.17–2.58)	0.5526		
Lethargy or loss of consciousness[b]	125/253 (49)	9/21 (43)	0.78 (0.32–1.87)	0.5759		
Dry mouth[b]	112/255 (44)	8/21 (38)	0.80 (0.33–1.96)	0.6304		
Fast breathing[b]	53/253 (21)	2/21 (10)	0.48 (0.12–1.85)	0.2875		
Undernutrition	69/255 (27)	4/21 (19)	0.69 (0.24–2.02)	0.4975		
Low or very low skin pinch	50/237 (21)	4/19 (21)	1.08 (0.36–3.22)	0.8931		
24–59 months of age						
Dysentery	46/116 (40)	22/29 (76)	4.55 (1.84–11.25)	0.0010	5.29 (2.05–13.66)	0.0006
Sunken eyes	43/116 (37)	8/29 (28)	0.67 (0.28–1.61)	0.3679		
Loss of skin turgor	20/116 (17)	2/29 (7)	0.43 (0.11–1.70)	0.2279		
Intravenous rehydration	34/116 (29)	5/29 (17)	0.54 (0.20–1.47)	0.2258		
Hospitalized	59/116 (51)	5/29 (17)	0.22 (0.08–0.59)	0.0026		
Vomiting 3 or more times per day[a]	38/116 (33)	3/29 (10)	0.27 (0.08–0.88)	0.0293		
Very thirsty[a]	93/116 (80)	22/29 (76)	0.75 (0.29–1.93)	0.5569		
Drank much less than usual[a]	11/116 (9)	1/29 (3)	0.48 (0.08–2.78)	0.4151		
Unable to drink[a]	9/116 (8)	3/29 (10)	1.49 (0.41–5.47)	0.5438		
Belly pain[a]	23/116 (20)	10/29 (34)	2.14 (0.89–5.14)	0.0883		
Fever[a]	40/115 (35)	11/29 (38)	1.16 (0.51–2.66)	0.7275		
Irritable or restless[a]	9/115 (8)	2/29 (7)	1.02 (0.24–4.37)	0.9796		
Decreased activity or lethargy[a]	58/115 (50)	10/29 (34)	0.53 (0.23–1.22)	0.1348		
Loss of consciousness[a]	2/116 (2)	1/29 (3)	2.41 (0.31–19.02)	0.4038		
Rectal straining[a]	9/115 (8)	5/29 (17)	2.52 (0.81–7.86)	0.1120		
Rectal prolapse[a]	0/116 (0)	1/29 (3)	12.26 (0.49–308.99)	0.1279	32.08 (1.18–869.71)	0.0394
Cough[a]	52/116 (45)	13/29 (45)	1.01 (0.45–2.25)	0.9900		
Difficulty breathing[a]	2/115 (2)	0/29 (0)	0.77 (0.04–16.46)	0.8669		
Convulsion[a]	8/116 (7)	1/29 (3)	0.67 (0.11–4.00)	0.6621		
Very thirsty[b]	94/116 (81)	22/29 (76)	0.71 (0.28–1.84)	0.4856		
Drinks poorly[b]	15/116 (13)	1/29 (3)	0.34 (0.06–1.94)	0.2266		
Sunken eyes[b]	45/116 (39)	8/29 (28)	0.62 (0.26–1.49)	0.2873		
Wrinkled skin[b]	19/116 (16)	2/29 (7)	0.45 (0.11–1.81)	0.2638		
Irritability or restless attitude[b]	3/116 (3)	2/29 (7)	2.95 (0.55–15.75)	0.2060		
Lethargy or loss of consciousness[b]	52/116 (45)	9/29 (31)	0.57 (0.24–1.33)	0.1946		
Dry mouth[b]	35/116 (30)	5/29 (17)	0.52 (0.19–1.41)	0.1962		
Fast breathing[b]	8/116 (7)	0/29 (0)	0.22 (0.01–3.86)	0.2977		
Undernutrition	8/116 (7)	0/29 (0)	0.22 (0.01–3.86)	0.2977		
Low or very low skin pinch	16/108 (15)	1/28 (4)	0.31 (0.05–1.72)	0.1782		

Notes: [a]Since began; [b]current.
Abbreviation: MSD, moderate-to-severe diarrhea.

associated with significant clinical features according to age, and the results from adjusted analysis are highlighted here. Dysentery, a classic symptom of shigellosis, was one of the most important features of diarrhea caused by *Shigella* compared to non-*Shigella*-MSD in both children aged 12–23 months (aOR =3.99; 95% CI: 153–10.39; *P*=0.0046) and 24–59 months (aOR =5.29; 95% CI: 2.05–13.66; *P*=0.0006). Moreover, the proportion of *Shigella* isolation was high among bloody diarrheal cases (20.1%; 32/158) compared to only 3.2% (24/758) for watery diarrhea, with the highest rate in children aged 24–59 months accounting with approximately one-third of all bloody diarrheal cases (Table 4). Fever was also an important symptom of *Shigella*-MSD in children from 12 to 23 months (aOR =3.02; 1.15–7.94; *P*=0.025) while rectal prolapse in 24–59 months (aOR =32.08; 1.18–869.71; *P*=0.0394). Irritability or restless attitude was an important sign for infants by crude analysis (OR =4.93; 1.10–22.9; *P*=0.0372).

Risk factors

The role of different risk factors for MSD was age-dependent, although most of the studied socioeconomic or hygienic

Table 4 Crude analysis of *Shigella*-associated diarrhea by severity (bloody diarrhea and watery diarrhea) in the different age groups (December 2007–November 2012)

Age groups	Watery diarrhea, n (%)	Bloody diarrhea, n (%)	OR (95% CI)	*P*-value
0–11 months	5/459 (1)	1/36 (3)	3.49 (0.56–21.93)	0.1823
12–23 months	12/222 (5)	9/54 (17)	3.52 (1.43–8.67)	0.0063
24–59 months	7/77 (9)	22/68 (32)	4.55 (1.84–11.25)	0.001

explanatory variables were not statistically significant (Table 5). Partial or exclusive breastfeeding was a remarkable protective factor against *Shigella* in younger children aged 0–11 months with MSD by crude analysis (OR =0.10; 0.02–0.68; *P*=0.0184). Protective factors in children aged 12–23 months included partial or exclusive breastfeeding (OR =0.12; 0.04–0.40; *P*=0.0006), mother as primary child caretaker (aOR =0.28; 0.09–0.87; *P*=0.0281), and giving stored water to child (aOR =0.17; 0.04–0.82; *P*=0.0267). In 24–59 months, water availability (not always per day) (aOR =0.32; 0.12–0.81; *P*=0.0167) and washing hands before preparing baby's food (aOR =0.28; 0.08–0.94; *P*=0.0394) were protective against shigellosis. Among controls (Table 6), relevant protective factors for acquiring *Shigella* included child primary caretaker for children aged 0–11 months (aOR =0.03; 0.00–0.87; *P*=0.0408), and washing hands before eating in children aged 12–23 months (aOR =0.05; 0.01, 0.52; *P*=0.0119).

Impact of antibiotic treatment on patient outcome

The impact of antibiotic treatment on patient outcome was assessed among children with known outcome (Table 7). Patient outcome was available for all of *Shigella*-diarrheal children while antibiotic treatment was administered in only 30.1% (22/73). The most administered antibiotics were ampicillin and gentamicin (12.3%; 9/73), followed by trimethoprim-sulfametoxazole (9.6%; 7/73), chloramphenicol (8.2%; 6/73), and nalidixic acid (6.9%; 5/73). Although high rates of resistance were found, patient improvement at discharge was much higher in children that received antibiotic treatment (63.0%; 17/27) compared to 37.0% (10/27), for those without any treatment (*P*<0.0001).

Discussion

In this prospective, case–control study, we demonstrated the association of *Shigella* spp. with MSD and LSD among children under 5 years in Manhiça District, a rural area of southern Mozambique. Its low detection rate in children without diarrhea and in cases of LSD confirms the high probability that whenever present, this pathogen causes a virulent disease, which could be explained in part by the low infectious dose of the bacterium (10–100 cells).[21] While an association between *Enterobacteriaceae* and *Giardia* has been reported,[22] in this study we showed that the prevalence of Giardia was independent of *Shigella* by adjusted analysis. In addition, *Giardia* was more common in controls while *Shigella* was mostly isolated in cases. The proportion of cases presenting with *Shigella* was similar with those reported in our previous study[12] and in other African countries.[3,23,24] Overall, the detection rate of *Shigella* was high during rainy season, which is similar to other studies.[25] As expected, the classical signs/symptoms of *Shigella*-MSD were mostly of dysentery accompanied with fever or rectal prolapse.[8] Irritability or restless attitude was also an importance feature in infants. The observation that *Shigella* may also be associated with less severe cases is a matter of concern, because the disease is mostly considered severe and frequently associated with dysentery. These findings expand the spectrum of clinical features associated with *Shigella* infection and demonstrate the importance of active surveillance for disease prevention. Another explanation could be related to health-seeking behavior, meaning that mothers or child caretakers are looking for assistance earlier before the disease becomes severe, as demonstrated in our community study about health care utilization and attitudes in cases of MSD.[26]

The finding that children who received antibiotic treatment was associated with a good outcome at the discharge, reinforcing the importance of antibiotic therapy for the management of shigellosis. Similar reports have shown that, with an effective antibiotic therapy, clinical improvement occurs within 48 hours,[27] resulting in a decreased risk of serious complications and death, shorter duration of symptoms, the elimination of *Shigella* from the stool, and subsequently decreased infection transmission.

MSD caused by *Shigella* was associated with a high OR compared to other significant pathogens in adjusted analysis, which may suggest its high pathogenicity as observed by the low detection rate in healthy children compared to other significant pathogens. In accordance with previous reports,[28]

Table 5 Risk factors of *Shigella*-associated moderate-to-severe diarrhea by crude and adjusted analysis

Variables	*Shigella* spp.		Crude analysis		Adjusted analysis	
	Negative, n (%)	Positive, n (%)	OR (95% CI)	*P*-value	Adjusted OR (95% CI)	*P*-value
0–11 months						
>6 months of age	259/489 (53)	5/6 (83)	3.26 (0.53–19.98)	0.2021		
Child sex (male)	294/489 (60)	4/6 (67)	1.19 (0.25–5.67)	0.8226		
Child primary caretaker (mother)	473/489 (97)	6/6 (100)	0.45 (0.02–8.38)	0.5948		
Caretaker formal education	135/487 (28)	3/6 (50)	2.60 (0.58–11.60)	0.2100		
Animals in compound	411/489 (84)	4/6 (67)	0.34 (0.07–1.64)	0.1806		
Water availability (not always per day)	256/488 (52)	4/6 (67)	1.63 (0.34–7.73)	0.5375		
Access to improved water	411/489 (84)	4/6 (67)	0.34 (0.07–1.64)	0.1806		
Give stored water to child	450/489 (92)	6/6 (100)	1.14 (0.06–20.61)	0.9294		
Treating water habit	54/489 (11)	1/6 (17)	2.18 (0.35–13.55)	0.4034		
Facility to dispose child's stool	248/486 (51)	2/6 (33)	0.53 (0.11–2.53)	0.4284		
Improved facility for household stool	43/488 (9)	0/6 (0)	0.79 (0.04–14.22)	0.8716		
Wash hands before eating	463/489 (95)	5/6 (83)	0.21 (0.03–1.33)	0.0974		
Wash hands before cooking	304/489 (62)	3/6 (50)	0.61 (0.14–2.71)	0.5152		
Wash hands before preparing baby's food	129/489 (26)	3/6 (50)	2.78 (0.62–12.42)	0.1796		
Wash hands after defecating	413/489 (84)	6/6 (100)	2.41 (0.13–43.14)	0.5513		
Wash hands after handling animals	12/489 (2)	0/6 (0)	2.94 (0.16–55.07)	0.4710		
Wash hands after cleaning child feces	127/489 (26)	2/6 (33)	1.58 (0.33–7.51)	0.5655		
Partial or exclusive breastfeeding	413/424 (97)	5/6 (83)	0.10 (0.02–0.68)	0.0184		
Lowest quintile of wealth index	110/484 (23)	2/6 (33)	1.88 (0.40–8.96)	0.4268		
12–23 months						
>18 months of age	75/255 (29)	10/21 (48)	2.18 (0.91–5.26)	0.0816		
Child sex (male)	147/255 (58)	10/21 (48)	0.67 (0.28–1.61)	0.3713		
Child primary caretaker (mother)	237/255 (93)	17/21 (81)	0.30 (0.10–0.95)	0.0398	0.28 (0.09–0.87)	0.0281
Caretaker formal education	58/253 (23)	4/21 (19)	0.86 (0.29–2.52)	0.7825		
Animals in compound	213/255 (84)	16/21 (76)	0.60 (0.22–1.66)	0.3217		
Water availability (not always per day)	140/255 (55)	11/21 (52)	0.90 (0.38–2.15)	0.8135		
Access to improved water	214/255 (84)	16/21 (76)	0.58 (0.21–1.61)	0.2962		
Give stored water to child	249/255 (98)	19/21 (90)	0.20 (0.04–0.94)	0.0412	0.17 (0.04–0.82)	0.0267
Treating water habit	14/255 (5)	2/21 (10)	2.14 (0.52–8.83)	0.2948		
Facility to dispose child's stool	207/253 (82)	18/21 (86)	1.18 (0.36–3.88)	0.7796		
Improved facility for household stool	15/255 (6)	0/21 (0)	0.36 (0.02–6.24)	0.4834		
Wash hands before eating	225/255 (88)	19/21 (90)	1.05 (0.27–4.15)	0.9390		
Wash hands before cooking	152/255 (60)	11/21 (52)	0.74 (0.31–1.78)	0.5055		
Wash hands before preparing baby's food	78/255 (31)	4/21 (19)	0.58 (0.20–1.69)	0.3203		
Wash hands after defecating	219/255 (86)	19/21 (90)	1.30 (0.33–5.07)	0.7083		
Wash hands after handling animals	4/255 (2)	0/21 (0)	1.30 (0.07–24.95)	0.8619		
Wash hands after cleaning child feces	59/255 (23)	7/21 (33)	1.71 (0.68–4.32)	0.2581		
Partial or exclusive breastfeeding	139/214 (65)	3/18 (17)	0.12 (0.04–0.40)	0.0006		
Lowest quintile of wealth index	55/255 (22)	2/21 (10)	0.46 (0.12–1.79)	0.2637		
24–59 months						
>42 months of age	25/116 (22)	3/29 (10)	0.47 (0.14–1.57)	0.2218		
Child sex (male)	68/116 (59)	19/29 (66)	1.31 (0.57–3.03)	0.5208		
Child primary caretaker (mother)	100/116 (86)	28/29 (97)	3.12 (0.56–17.46)	0.1955		
Caretaker formal education	23/115 (20)	11/29 (38)	2.45 (1.03–5.81)	0.0424		
Animals in compound	95/116 (82)	24/29 (83)	1.00 (0.36–2.83)	0.9957		
Water availability (not always per day)	60/116 (52)	7/29 (24)	0.31 (0.13–0.77)	0.0113	0.32 (0.12–0.81)	0.0167
Access to improved water	95/116 (82)	25/29 (86)	1.28 (0.42–3.86)	0.6661		
Give stored water to child	109/115 (95)	27/29 (93)	0.65 (0.14–2.98)	0.5819		
Treating water habit	5/116 (4)	3/29 (10)	2.68 (0.66–10.92)	0.1696		
Facility to dispose child's stool	110/116 (95)	28/29 (97)	1.12 (0.18–6.92)	0.9048		
Improved facility for household stool	13/116 (11)	4/29 (14)	1.35 (0.43–4.28)	0.6069		
Wash hands before eating	106/116 (91)	27/29 (93)	1.08 (0.26–4.58)	0.9122		
Wash hands before cooking	70/116 (60)	19/29 (66)	1.22 (0.53–2.83)	0.6346		
Wash hands before preparing baby's food	34/116 (29)	3/29 (10)	0.32 (0.10–1.03)	0.0563	0.28 (0.08–0.94)	0.0394
Wash hands after defecating	101/116 (87)	24/29 (83)	0.68 (0.23–1.98)	0.4796		
Wash hands after handling animals	4/116 (3)	1/29 (3)	1.32 (0.20–8.74)	0.7763		
Wash hands after cleaning child feces	23/116 (20)	6/29 (21)	1.10 (0.41–2.93)	0.8480		
Partial or exclusive breastfeeding	18/100 (18)	4/20 (20)	1.22 (0.38–3.87)	0.7404		
Lowest quintile of wealth index	31/115 (27)	7/29 (24)	0.89 (0.36–2.25)	0.8121		

Table 6 Risk factors for *Shigella* infection in controls by crude and adjusted analysis

Variables	*Shigella* spp.		Crude analysis		Adjusted analysis	
	Negative, n (%)	Positive, n (%)	OR (95% CI)	*P*-value	Adjusted OR (95% CI)	*P*-value
0–11 months						
>6 months of age	595/1046 (57)	1/1 (100)	2.27 (0.09–55.97)	0.6151		
Child sex (male)	629/1046 (60)	1/1 (100)	1.99 (0.08–48.96)	0.6738		
Child primary caretaker (mother)	1035/1046 (99)	1/1 (100)	0.03 (0.00–0.86)	0.0404	0.03 (0.00–0.87)	0.0408
Caretaker formal education	313/1044 (30)	1/1 (100)	7.00 (0.28–172.30)	0.2338		
Animals in compound	897/1045 (86)	1/1 (100)	0.50 (0.02–12.24)	0.6684		
Water availability (not always per day)	333/1046 (32)	0/1 (0)	0.71 (0.03–17.55)	0.8361		
Access to improved water	888/1046 (85)	0/1 (0)	0.06 (0.00–1.47)	0.0844		
Give stored water to child	829/1046 (79)	0/1 (0)	0.09 (0.00–2.15)	0.1360		
Treating water habit	66/1046 (6)	0/1 (0)	4.91 (0.20–121.81)	0.3310		
Facility to dispose child's stool	656/1046 (63)	1/1 (100)	1.78 (0.07–43.91)	0.7231		
Improved facility for household stool	79/1046 (8)	0/1 (0)	4.06 (0.16–100.39)	0.3924		
Wash hands before eating	980/1046 (94)	1/1 (100)	0.20 (0.01–5.04)	0.3310		
Wash hands before cooking	855/1046 (82)	0/1 (0)	0.07 (0.00–1.84)	0.1124		
Wash hands before preparing baby's food	627/1046 (60)	1/1 (100)	2.01 (0.08–49.35)	0.6702		
Wash hands after defecating	765/1046 (73)	1/1 (100)	1.10 (0.04–27.16)	0.9521		
Wash hands after handling animals	421/1046 (40)	0/1 (0)	0.49 (0.02–12.17)	0.6667		
Wash hands after cleaning child feces	642/1046 (61)	1/1 (100)	1.89 (0.08–46.48)	0.6972		
Partial or exclusive breastfeeding	1018/1039 (98)	1/1 (100)	0.06 (0.00–1.60)	0.0939		
Lowest quintile of wealth index	194/1045 (19)	0/1 (0)	1.46 (0.06–35.96)	0.8172		
12–23 months						
>18 months of age	200/627 (32)	0/2 (0)	0.43 (0.02–8.92)	0.5828		
Child sex (male)	373/627 (59)	2/2 (100)	3.41 (0.16–71.26)	0.4294		
Child primary caretaker (mother)	598/627 (95)	2/2 (100)	0.25 (0.01–5.25)	0.3695		
Caretaker formal education	172/625 (28)	1/2 (50)	2.63 (0.27–25.45)	0.4039		
Animals in compound	555/627 (89)	2/2 (100)	0.65 (0.03–13.73)	0.7836		
Water availability (not always per day)	216/627 (34)	2/2 (100)	9.50 (0.45–198.84)	0.1467		
Access to improved water	521/627 (83)	1/2 (50)	0.20 (0.02–1.98)	0.1707		
Give stored water to child	583/625 (93)	2/2 (100)	0.36 (0.02–7.71)	0.5166		
Treating water habit	26/627 (4)	0/2 (0)	4.54 (0.21–96.93)	0.3327		
Facility to dispose child's stool	600/626 (96)	2/2 (100)	0.22 (0.01–4.71)	0.3333		
Improved facility for household stool	27/627 (4)	0/2 (0)	4.37 (0.20–93.17)	0.3451		
Wash hands before eating	596/627 (95)	1/2 (50)	0.05 (0.01–0.52)	0.0119	0.05 (0.01–0.52)	0.0119
Wash hands before cooking	522/627 (83)	2/2 (100)	1.01 (0.05–21.18)	0.9951		
Wash hands before preparing baby's food	386/627 (62)	0/2 (0)	0.12 (0.01–2.61)	0.1801		
Wash hands after defecating	454/627 (72)	2/2 (100)	1.91 (0.09–39.96)	0.6770		
Wash hands after handling animals	254/627 (41)	0/2 (0)	0.29 (0.01–6.14)	0.4294		
Wash hands after cleaning child feces	367/627 (59)	0/2 (0)	0.14 (0.01–2.97)	0.2079		
Partial or exclusive breastfeeding	443/621 (71)	2/2 (100)	2.01 (0.10–42.13)	0.6522		
Lowest quintile of wealth index	135/627 (22)	1/2 (50)	3.63 (0.38–35.22)	0.2654		
24–59 months						
>42 months of age	51/302 (17)	0/1 (0)	1.63 (0.07–40.52)	0.7664		
Child sex (male)	201/302 (67)	0/1 (0)	0.17 (0.01–4.16)	0.2759		
Child primary caretaker (mother)	268/302 (89)	1/1 (100)	0.39 (0.02–9.65)	0.5618		
Caretaker formal education	74/302 (25)	0/1 (0)	1.02 (0.04–25.37)	0.9892		
Animals in compound	252/302 (83)	1/1 (100)	0.60 (0.02–14.94)	0.7555		
Water availability (not always per day)	103/302 (34)	0/1 (0)	0.64 (0.03–15.91)	0.7870		
Access to improved water	254/302 (84)	1/1 (100)	0.57 (0.02–14.24)	0.7332		
Give stored water to child	277/302 (92)	1/1 (100)	0.28 (0.01–6.94)	0.4337		
Treating water habit	9/302 (3)	0/1 (0)	10.30 (0.39–269.66)	0.1616		
Facility to dispose child's stool	298/301 (99)	1/1 (100)	0.04 (0.00–1.02)	0.0515		
Improved facility for household stool	24/302 (8)	0/1 (0)	3.79 (0.15–95.51)	0.4185		
Wash hands before eating	286/302 (95)	1/1 (100)	0.17 (0.01–4.41)	0.2880		
Wash hands before cooking	232/302 (77)	1/1 (100)	0.91 (0.04–22.58)	0.9539		
Wash hands before preparing baby's food	156/302 (52)	0/1 (0)	0.31 (0.01–7.72)	0.4768		
Wash hands after defecating	210/302 (70)	0/1 (0)	0.15 (0.01–3.63)	0.2408		
Wash hands after handling animals	122/302 (40)	1/1 (100)	4.42 (0.18–109.40)	0.3640		
Wash hands after cleaning child feces	161/302 (53)	1/1 (100)	2.63 (0.11–65.04)	0.5550		
Partial or exclusive breastfeeding	8/279 (3)	0/1 (0)	10.65 (0.40–280.87)	0.1566		
Lowest quintile of wealth index	53/302 (18)	0/1 (0)	1.55 (0.06–38.68)	0.7879		

Table 7 Impact of antibiotic treatment for *Shigella*-diarrhea in children outcome when leaving the hospital from December 2007 to November 2012

Outcomes	Antibiotic treatment, n (%)			
	No	Yes	Total	*P*-value
Improved	10/27 (37)	17/27 (63)	27	<0.0001
Not improved	41/46 (89.1)	5/46 (10.8)	46	
Total	51/73 (69.8)	22/73 (30.1)	73	
Antibiotics administered	**Proportion, n (%)**		**Resistance, n (%)**	
Trimethoprim-sulfametoxazole	7/73 (9.6)		62/67 (92.5)	
Gentamicin	9/73 (12.3)		0	
Chloramphenicol	6/73 (8.2)		36/67 (53.7)	
Amoxicillin clavulanic acid	4/73 (5.5)		3/67 (4.5)	
Ampicillin	9/73 (12.3)		34/67 (50.7)	
Nalidixic acid	5/73 (6.9)		0	
Ciprofloxacin	1/73 (1.4)		0	

we found a low risk of *Shigella*-associated diarrhea in infancy and a steady increase through the second year of life. One likely explanation is that the partial or exclusive breastfeeding, which was present in 97% of infants (<12 months) of age and in 61% of children under 2 years in our study, neutralized the infectious inoculum and prevented disease. The strong association between MSD caused by *Shigella* and children over 2 years of age has been attributed to a decreased exposure to breast milk and an increased exposure to sources of *Shigella* infection such as hands contact in the environment as children become more active.[29,30] In addition, the benefits of human milk for infant health have been recognized as an important source of bacteria that may contribute to neonatal gastrointestinal colonization leading to immune response development and possible infection prevention. Thus, the microbiota of breast-fed infants is considered the gold standard in terms of a healthy infant gastrointestinal microbiota.[31]

In addition, as children wean, they are exposed to an increased array of food and water that could serve as sources of *Shigella,* thus increasing the probability of getting infected.[32] Therefore, this study also sought to quantify the effects of several socioeconomic indicators on the relative increased risk for acquiring *Shigella* in children presenting MSD. Overall, the role of the assessed risk factors for *Shigella*-MSD was age-related, possibly due to differences in the degree of exposure of the studied population. The presence of broader protective factors for diarrhea caused by *Shigella* in the present study could be attributed to an improved health-seeking behavior among the cases and improvement in accessibility of health services over the years. The population leaving in Manhiça district is under monitoring through HDSS since 1996, and is thus exposed to several interventions that contribute to health education that may influence the health-seeking behavior. This may explain why socioeconomic characteristics such as formal education were less relevant in the present study. Considering these variations, the importance of children primary caretaker (mother), giving stored water to child, water availability (not always per day), and washing hands before preparing baby's food in preventing diarrhea caused by *Shigella* is highlighted.

The importance of hand-washing practices in preventing diarrheal illness is supported by a number of studies.[33,34] A systematic review of several studies estimated that appropriate hand washing with soap could reduce the risks of severe intestinal infections and shigellosis up to 48% and 59%, respectively.[34] The fact that stored water consumption was protective for *Shigella* infection leads us to believe that the hygienic conditions for water transportation and the containers where the water is stored are adequate or properly cleaned, because water contamination can occur at any stage from the source to the point of use. Conversely, unavailability of water throughout the day was protective for *Shigella* infection and may reflect the rational use of scarce water at the household level, or be linked to the hypothesis that the lack of water can interrupt the chain of transmission of some waterborne transmitted enteropathogens like *Shigella* as families are less exposed due to the limited amount of water that they have. The prevalence of malnutrition in both *Shigella*-associated MSD and in non-*Shigella* MSD highlights the vicious cycle of malnutrition and diarrhea. While malnutrition makes children more vulnerable to diarrheal infections due to its negative effect on the barrier protection leading to deterioration of the immune system, chronic infections can contribute to long-term gut damage and preventing nutrient absorption.[35]

Conclusion

Shigella is a significant pathogen associated with MSD in Mozambican children from 1 to 5 years. The presence of cases of LSD caused by *Shigella* is a matter of concern and may suggest the need to expand the clinical feature of the disease. The presence of dysentery remains an important clinical feature of *Shigella*-associated diarrhea. The study reinforces the recommendations from the WHO on the use of antibiotic treatment for the management of shigellosis while vaccines to prevent disease are not available. As demonstrated in this study, promoting awareness of good personal hygiene and a safe water storage is of great importance in prevention and control of the disease. Moreover, continuous surveillance studies are needed to track changes over time.

Acknowledgments

The authors thank all children and their parents for participating in the surveillance. Thanks to clinicians, nurses, and other staff from CISM and Manhiça District Hospital for collecting and processing the data. Thanks to the district health authorities and the Ministry of Health for their collaboration in the research activities ongoing in Manhiça District.

This study was part the GEMS study funded by the Bill and Melinda Gates Foundation. CISM receives core funds from Spanish Agency for International Cooperation and Development (AECID). Delfino Vubil received a fellowship from Fundação Calouste Gulbenkian – Programa Gulbenkian Parcerias para o Desenvolvimento (www.gulbenkian.pt).

Disclosure

The authors report no conflicts of interest in this work.

References

1. Das JK, Tripathi A, Ali A, Hassan A, Dojosoeandy C, Bhutta ZA. Vaccines for the prevention of diarrhea due to cholera, shigella, ETEC and rotavirus. *BMC Public Health*. 2013;13 Suppl 3:S11.
2. Levine MM, Kotloff KL, Nataro JP, Muhsen K. The Global Enteric Multicenter Study (GEMS): impetus, rationale, and genesis. *Clin Infect Dis*. 2012;55 Suppl 4:S215–S224.
3. Bonkoungou IJ, Haukka K, Österblad M, et al. Bacterial and viral etiology of childhood diarrhea in Ouagadougou, Burkina Faso. *BMC Pediatr*. 2013;13:36.
4. Tian L, Zhu X, Chen Z, et al. Characteristics of bacterial pathogens associated with acute diarrhea in children under 5 years of age: a hospital-based cross-sectional study. *BMC Infect Dis*. 2016;16(1):1–8.
5. Kotloff KL, Winickoff JP, Ivanoff B, et al. Global burden of Shigella infections: implications for vaccine development and implementation of control strategies. *Bull World Health Organ*. 1999;77(8):651–666.
6. Bohles N, Busch K, Hensel M. Vaccines against human diarrheal pathogens: current status and perspectives. *Hum Vaccines Immunother*. 2014;10:1522–1535.
7. Aragón TJ, Vugia DJ, Shallow S, et al. Case-control study of shigellosis in San Francisco: the role of sexual transmission and HIV infection. *Clin Infect Dis*. 2007;44(3):327–334.
8. Sur D, Ramamurthy T, Deen J, Bhattacharya SK. Shigellosis: challenges & management issues. *Indian J Med Res*. 2004;120(5):454–462.
9. Traa BS, Walker CL, Munos M, Black RE. Antibiotics for the treatment of dysentery in children. *Int J Epidemiol*. 2010;39 Suppl 1:i70–i74.
10. El Bushra HE, Bin Saeed AA. Intrafamilial person-to-person spread of bacillary dysentery due to Shigella dysenteriae in southwestern Saudi Arabia. *East Afr Med J*. 1999;76(5):255–259.
11. Nhampossa T, Mandomando I, Acacio S, et al. Diarrheal disease in rural Mozambique: burden, risk factors and etiology of diarrheal disease among children aged 0-59 months seeking care at health facilities. *PLoS One*. 2015;10(5):e0119824.
12. Mandomando I, Sigaúque B, Vallès X, et al. Epidemiology and clinical presentation of shigellosis in children less than five years of age in rural Mozambique. *Pediatr Infect Dis J*. 2007;26(11):1059–1061.
13. Kotloff KL, Blackwelder WC, Nasrin D, et al. The Global Enteric Multicenter Study (GEMS) of diarrheal disease in infants and young children in developing countries: epidemiologic and clinical methods of the case/control study. *Clin Infect Dis*. 2012;55(Suppl 4):S232–S245.
14. González R, Munguambe K, Aponte J, et al. High HIV prevalence in a southern semi-rural area of Mozambique: a community-based survey. *HIV Med*. 2012;13(10):581–588.
15. Sacarlal J, Nhacolo AQ, Sigaúque B. A 10 year study of the cause of death in children under 15 years in Manhiça, Mozambique. *BMC Public Health*. 2006;3002:1–10.
16. Sacoor C, Nhacolo A, Nhalungo D, et al. Profile: Manhiça Health Research Centre (Manhiça HDSS). *Int J Epidemiol*. 2013;42(5):1309–1318.
17. Kotloff KL, Nataro JP, Blackwelder WC, et al. Burden and aetiology of diarrhoeal disease in infants and young children in developing countries (the Global Enteric Multicenter Study, GEMS): a prospective, case-control study. *Lancet*. 2013;382(9888):209–222.
18. Panchalingam S, Antonio M, Hossain A, et al. Diagnostic microbiologic methods in the GEMS-1 case/control study. *Clin Infect Dis*. 2012;55(Suppl 4):S294–S302.
19. Firth D. Bias reduction of maximum likelihood estimates. *Biometrika*. 1993;80:27–38.
20. Heinze G, Schemper M. A solution to the problem of separation in logistic regression. *Stat Med*. 2002;21(16):2409–2419.
21. Mattock E, Blocker AJ. How do the virulence factors of Shigella work together to cause disease? *Front Cell Infect Microbiol*. 2017;7:1–24.
22. Zerpa R, Huicho L. Intestinal coinfection with numerous Giardia trophozoites and Vibrio cholerae in hospitalized children with watery diarrhea. *Wilderness Environ Med*. 1995;6(2):167–172.
23. Nitiema LW, Nordgren J, Ouermi D, et al. Burden of rotavirus and other enteropathogens among children with diarrhea in Burkina Faso. *Int J Infect Dis*. 2011;15(9):e646–e652.
24. Moyo SJ, Gro N, Matee MI, et al. Age specific aetiological agents of diarrhoea in hospitalized children aged less than five years in Dares Salaam, Tanzania. *BMC Pediatr*. 2011;11:19.
25. Lee HS, Ha Hoang TT, Pham-Duc P, et al. Seasonal and geographical distribution of bacillary dysentery (shigellosis) and associated climate risk factors in Kon Tum Province in Vietnam from 1999 to 2013. *Infect Dis Poverty*. 2017;6(1):1–11.
26. Nhampossa T, Mandomando I, Acacio S, et al. Health care utilization and attitudes survey in cases of moderate-to-severe diarrhea among children ages 0-59 months in the District of Manhica, southern Mozambique. *Am J Trop Med Hyg*. 2013;89(1 Suppl):41–48.
27. World Health Organisation. *Guidelines for the control of shigellosis, including epidemics due to Shigella dysenteriae type 1*. Geneva: World Health Organization, 2005. Available from: http://apps.who.int/iris/bitstream/10665/43252/1/924159330X.pd. Accessed October 3, 2018.
28. Abu-Elyazeed RR, Wierzba TF, Frenck RW, et al. Epidemiology of Shigella-associated diarrhea in rural Egyptian children. *Am J Trop Med Hyg*. 2004;71(3):367–372.

29. Clemens JD, Stanton B, Stoll B, Shahid NS, Banu H, Chowdhury AK. Breast feeding as a determinant of severity in shigellosis. Evidence for protection throughout the first three years of life in Bangladeshi children. *Am J Epidemiol*. 1986;123(4):710–720.
30. Ahmed F, Clemens JD, Rao MR, Sack DA, Khan MR, Haque E. Community-based evaluation of the effect of breast-feeding on the risk of microbiologically confirmed or clinically presumptive shigellosis in Bangladeshi children. *Pediatrics*. 1992;90(3):406–411.
31. Murphy K, Curley D, O'Callaghan TF, et al. The composition of human milk and infant faecal microbiota over the first three months of life: a pilot study. *Sci Rep*. 2017;7:40597.
32. Lindsay B, Saha D, Sanogo D, et al. Association between shigella infection and diarrhea varies based on location and age of children. *Am J Trop Med Hyg*. 2015;93(5):918–924.
33. Maponga BA, Chirundu D, Gombe NT, Tshimanga M, Shambira G, Takundwa L. Risk factors for contracting watery diarrhoea in Kadoma City, Zimbabwe, 2011: a case control study. *BMC Infect Dis*. 2013;13:567.
34. Curtis V, Cairncross S. Effect of washing hands with soap on diarrhoea risk in the community: a systematic review. *Lancet Infect Dis*. 2003;3(5):275–281.
35. Rodríguez L, Cervantes E, Ortiz R. Malnutrition and gastrointestinal and respiratory infections in children: a public health problem. *Int J Environ Res Public Health*. 2011;8(4):1174–1205.

Infection and Drug Resistance Dovepress

Publish your work in this journal

Infection and Drug Resistance is an international, peer-reviewed open-access journal that focuses on the optimal treatment of infection (bacterial, fungal and viral) and the development and institution of preventive strategies to minimize the development and spread of resistance. The journal is specifically concerned with the epidemiology of antibiotic resistance and the mechanisms of resistance development and diffusion in both hospitals and the community. The manuscript management system is completely online and includes a very quick and fair peer-review system, which is all easy to use. Visit http://www.dovepress.com/testimonials.php to read real quotes from published authors.

Submit your manuscript here: https://www.dovepress.com/infection-and-drug-resistance-journal