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Aetiology of Acute Gastro-Enteritis in Children at Saint Camille Medical Centre, Ouagadougou, Burkina Faso

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Abstract: The present study aims at identifying the infectious agents responsible for child Acute Gastro-Enteritis (AGE) in Ouagadougou. From May 5 2006 to June 22 2008, 648 children aged from 2 to 41 months, with at least an average of 3 loose stools per day have been enrolled for coproculture, parasitology and virology test. Among them, 34 (5.25%) were HIV seropositive. A single sample of faeces from each child was used to identify enteropathogens. An infectious aetiology was identified in 41.20% of cases. The pathogenic agents detected as responsible for the AGE are: Rotavirus 21.1%; Adenovirus 1.9%; Giardia 7.6% *Entamoeba*; 1.08%; entero-pathogenic *E. coli* 41.7%; Salmonella 3.40%; Shigella 1.85% and Yersinia 1.70%. Conclusion: Therefore, these AGE etiologic agents constitute a problem of public health in Burkina Faso. Their control for the child would require: (1) a regular paediatric and clinical follow up; (2) health education of the population for food hygiene and (3) in case of absence of HIV infection in the mother, a promotion of exclusive breast-feeding up to the age of 4 months.

Key words: Acute-gastro-enteritis, diarrhoea, rotavirus, giardia intestinalis, *E. coli* EPEC, salmonella

INTRODUCTION

Burkina Faso is a West African country bordering with Mali in the North, Niger in the East and Cote d'Ivoire, Ghana, Togo and Benin in the South. One of the major problems in this country is emaciation in children less than 5 years old, caused by Acute Gastro-Enteritis (AGE), which generally entails periods of dehydrating diarrhoea and malnutrition (Simpore *et al*., 2005; Pignatelli *et al*., 2000). AGE is responsible for 30 to 50% of the cases of acute paediatric diarrhoea in poor countries (Giordano *et al*., 2001). Acute diarrhoea constitutes one of the major causes of child morbidity and mortality and accounts for an estimated yearly 2 million deaths among children less than 5 years old in the world (Orlandi *et al*., 2006; Luz *et al*., 2005). About 130 million children suffer from diarrhoea caused by Rotavirus (RV) every year and among them, 18 million develop moderate to acute dehydration, resulting in 418.000 to 520.000 deaths (Luz *et al*., 2005; Ouermi *et al*., 2007). In Burkina Faso, the incidence of diarrhoea is 6 to 8 episodes per child per year among children less than 5 years old. (Ouermi *et al*., 2007; Sanou *et al*., 1999). The immediate causes are often infectious and include a variety of pathogenic micro organisms, namely viruses, bacteria and protozoa (Giordano *et al*., 2001). Nowadays, paediatric infections in developing countries are increasing due to: (1) lack in alimentary hygiene for children who crawl on the ground, drink dirty water and eat sand (Ouermi *et al*., 2007); (2) HIV/AIDS infection, which implies the lowering of the immune system, favouring various co-infections (Mbaye *et al*., 2005) and (3) absence of hygiene in public hospitals, causing transversal transmission of pathogenic agents and nosocomial infections.

The objective of the present study was to identify causative agents of AGE and to assess their prevalence.

MATERIALS AND METHODS

From May 5, 2006 to June 22, 2008, 648 samples of faeces were collected from children suffering from acute diarrhoea, with the occurrence of at least 3 loose or liquid faeces or stools per day, with or without vomiting. The specimen were cultured and analysed in the biomedical

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laboratory at Saint Camille, Ouagadougou. Among them, 34 (5.25%) were HIV seropositive. Most of these children came from poor peripheral neighbourhoods of Ouagadougou, where food hygiene is not well practiced. They were all followed up by our ambulatory team and a questionnaire was filled up during the medical consultation, specifying the age and sex. The same day, anthropometric measures such as weight and height were taken. For each sample of faeces, three types of testing were carried out:

- **Detection of intestinal parasites:** The faeces were collected early in the morning in plastic bags and each sample was submitted to a microscope test (to detect the presence of blood, mucus or adult worms) within 30 min after each emission of faeces. The standard parasitological test included a direct analysis of fresh stools, a technique of concentration type WILLIS and the use of LUGOL *E. histolytica* and *Giardia intestinalis*' cysts identification (Arslan *et al*., 2008)
- **Coproculture:** Each sample was cultured in a specific medium in conformity with the current norms, such as the salmonella-shigella medium (SS) and the Thiosulfate Citrate Bile Sucrose medium (TCBS) to isolate *V. cholera*. The EMB medium was used for the detection of E. coli EPEC for children and gelose Yiersinia CIN to isolate Yiersinia enterocolitica. The identification of bacteria was carried out by an observation of colonies, then morphological features were observed at the microscope and results were confirmed by the API 20 system (bioMérieux, France) (Dortet *et al*., 2006)
- Research on viruses was carried out with Adeno-Strip and Rota-Strip protocols of tests of inter medial srl. Via A Genovesi 13-80010 Villaricca NA, which are kits that enable a detection of Adenovirus enteritis (ADE) and Rotavirus (RV) in faeces with the use of an immunochromatographic technique on membrane (Ouermi *et al*., 2007)

The HIV test: after their mother's knowledgeable consent, 5 mL of blood was taken from each child suffering from AGE and put into an EDTA tube for the HIV serology test. For this analysis, two quick tests were sequentially used: Determine and Genie-II to detect HIV 1 and HIV 2, as previously described by Koblavi-Deme *et al.* (2001). After two quick tests with discordant results, a third test was used. In such cases the samples were tested with the ELISA technique, using the Abbott-IMX System (Abbott laboratories, N. Chicago), in order to confirm or exclude a result of HIV infection.

In addition, children's anthropometric measures (weight and height) were taken to determine their state of dehydration and malnutrition by Z-scores with the use of international references on the concerned population defined by the US National Center for Health Statistics (NCHS) (Simpore *et al*., 2005). The children were then classified according to the Z-score (or SD-score: Standard Deviation score). This system of classification according to Z-score recommended by the WHO and UNICEF services, uses weight/height (WHZ) nutritional indices, weight/age (WAZ) and this is done according to the following criteria: Z-score inferior to -3: corresponds to acute malnutrition; Z-score between -3 and -2: means a moderate malnutrition and finally a Z-score superior to -2 corresponds to a good nutrition (Simpore *et al*., 2005).

Ethical aspect: This study was approved by the Ethical Committee of Saint Camille Medical Centre. We also got free and knowledgeable oral consent of children's mothers.

Data processing: Statistic analyses carried out with software Epi-info version 6 and SPSS version 12. The value of $p \leq 0.05$ has been considered as significant.

RESULTS

The studies were effected on 648 children suffering from AGE and among them 34 were infected with HIV/AIDS. Table 1 shows the classification and anthropometrical features of the concerned population through a study according to age. Let us specify that in our sample there was a significant statistical difference between the two sexes as for averages in age ($p<0.001$).

Because of the severe dehydrating diarrhoea provoked by AGE, among the studied children, 405 over 648 (62.50%) presented an insufficient weight for their age and 335 children over 648 (51.70%) had an insufficient weight for their height (Table 2).

Table 1: Averages of age, weight and height of the children according to the total number of each age class

Averages				
Age class (months)	No.	Age (months)	Weight (kg)	Height (cm)
2<X<10	292	7.01±2.28	5.98±1.67	65.58±5.00
11<X<20	247	15.11±2.23	7.57±1.20	72.60±3.59
21<X<30	82	23.07±1.96	8.91±1.68	79.28±5.30
X>30	27	36.11±3.17	11.37±0.67	92.63±2.22
Total	648	13.34±7.62	7.18±2.00	71.11±7.89

Table 2: No. of children (648) per Z-score value: height/age (HAZ), weight/height (WHZ) and weight/age (WAZ), correlated with nutritional status

P (X= x)	HAZ	WHZ	WAZ
X<-3.00	107/648 (16.5%)	69/648 (13.7%)	207/648 (31.9%)
-2.99<X<-2.00	60/648 (9.3%)	246/648 (38.0%)	198/648 (30.6%)
X>-2	481/648 (74.2%)	313/648 (74.2%)	243/648 (37.5%)

Table 3: Frequency of rotavirus, adenovirus, HIV and intestinal parasites

					Prevalence of the various pathogens (%)						
Age class (months)	Number	RV	AdE	HIV	*G. Intestinalis*	*E. histolytca*	*E. coli*	*Salmonella*	*Shigella*	*Yersinia*	*V. cholera*
2<X<10	292	24.7	0.0	5.1	0.0	0.0	1.0	0.0	0.0	0.0	0.0
11<X<20	247	21.4	1.6	5.3	12.6	1.2	4.1	5.5	2.4	1.2	0.0
21<X<30	82	13.4	8.5	6.1	15.8	2.4	10.3	6.1	4.9	8.5	0.0
X>30	27	3.7	3.7	3.7	18.5	7.4	22.2	11.1	7.4	3.7	0.0
Total	648	21.1	1.8	5.2	7.6	1.1	4.2	3.4	1.9	1.7	0.0

NS: Not significant, Rotavirus 1¬2: p = 0.380 (NS); 1¬3: p = 0.030; 1¬4: p = 0.013; 2¬3: p = 0.111 (NS); 2¬4: p = 0.028; 3¬4: p = 0.297 SNS

Table 4: Serology of HIV-1 correlated with various co-infections and anthropometric parameters

		Prevalence of the various pathogens (%)						
		RV	AdE	AdE	*Giardia interstinalis*	*Entamoeba histolytica*	*Salmonella* sp.	*Shigella*
HIV$^+$	34	13/34	1/34	3/34	1/34	15/34	8/34	11/34
		38.2%	2.9%	8.8%	2.9%	44.1%	23.5%	32.4%
HIV$^-$	614	124/614	11/614	46/614	24/614	7/614	4/614	16/614
		20.2%	1.8%	7.5%	3.9%	1.1%	0.7%	2.6%
Total	648	137/648	12/648	49/648	25/648	22/648	12/648	7/648
		21.1%	1.9%	7.6%	3.8%	3.4%	1.9%	4.2%
χ^2 Test		p = 0.012	-		Yates's χ^2 Test p = 0.96^{NS}	Yates's χ^2 Test p = 0.86^{NS}	Yates's χ^2 Test p<0.001	-

	Prevalence of hte various pathogens (%)		Averages of anthropometric parameters				
	E. coli EPEC	*Yersinia*	Age	Height	Weight	WHZ	WAZ
HIV$^+$	Apr-34	Apr-34	11.18±7.11	69.10±6.20	6.29±1.17	-2.85±0.99	-3.63±1.01
	11.8%	11.8%					
HIV$^-$	7/614	7/614	13.46±7.63	71.22±7.96	7.21±2.02	-2.18±1.97	-3.16±1.21
	1.1%	1.1%					
Total	11/648	11/648	13.34±7.62	71.11±7.89	7.16±1.99	-2.21±1.96	-3.19±1.26
	1.7%	1.7%					
χ^2 Test	p<0.001	-	p = 0.09^{NS}	p = 0.01	p = 0.13^{NS}	p = 0.05	p = 0.03

^{NS}: Not significant

The biological test enabled us to identify the aetiological agents of AGE: parasites (*Giardia intestinalis*, *Entamoeba histolytica*), bacteria (*Escherichia coli* EPEC, *Salmonella*, *Shigella*, *Yersinia*) and viruses (Rotavirus and Adenovirus) (Table 3). Other parasites (*Hyménolopis nana*, *Ténia intestinalis*, *Strongyloids stercoralis*) and bacteria (*Klebsiella pneumoniae* and *Shigella flexneri*) non inductive pathogens of AGE have been isolated. Table 4 shows the HIV-1 serology association with various co-infections.

DISCUSSION

Through this study, we notice that children from 2 to 10 months of age constitute the most representative age class in the sample with a ratio of 292/648 (Table 1). Sanou *et al*. (1999) in their study, carried out at Yalgado Ouédraogo Hospital in Burkina Faso and Orlandi *et al*. (2006) in their research carried out at Porto Velho in Brazil, have respectively presented frequencies of diarrhoea of 55.7 and 53.3% for children of this same age class. During the first months of the child's life, the variety of the repertory of antibodies against infectious agents is limited (Weitkamp *et al*., 2003). It is precisely at this period that the child's own immunity develops progressively and that a lowering of maternal antibodies occurs. It is at the interface between the development of these anti-bodies and the loss of maternal immunity that the crucial moment of the foster child's immunological vulnerability is situated (Sanou *et al*., 1999). In rural areas as well as in town, when the infant starts crawling on hands and knees, he eats some earth, touches objects around him and puts everything into his mouth: it is the period of high contact with pathogenous microorganisms (Ouermi *et al*., 2007). In this vision, there is a strong link between overcrowding, poverty and high prevalence of diarrhoea in African cities. On the other hand, for the age class between 11 and 20 months, the frequency of diarrhoea could be related to contaminants in water and in solid foods progressively introduced in the child's diet, since breastfeeding in Burkina Faso is almost never exclusive.

Virology: on the entire sample, we could identify 137 (21.1%) seropositive children with the RV test (Table 4). Our values are superior to those obtained, respectively by Sanou *et al*. (1999) in Ouagadougou (14.4%), Olesen *et al*. (2005) in Danemark (13.2%) and Cardoso *et al*. (2003) at Goiânia (in Brazil) (14.4%). However, similar rates have been obtained by Mala in Burkina Faso (1993) (21.5%) and Ouermi *et al*. (2007) in Ouagadougou (22.7%) as well as Fodha *et al*. (2006) in Tunisia (20.0%). The same value remains inferior to those

of Kim *et al.* (1990) in Korea (68%), of Giordano *et al.* (2001) at Cordoba in Argentina (35.3%) and Armah *et al.* (2003) in Ghana (40.5%). These data show that RV has an incidence which varies from one country to another and inside the same country, but they remain the principal etiologic agent in viral AGE everywhere in the world (Gerba *et al.*, 1996). In this study, the frequency of RV infection significantly decreases according to age classes 2-10 months → 21-30 months ($p = 0.030$); 2-10 months → 31-41 months ($p = 0.013$) and from 11-20 months → 31-41 months ($p = 0.028$) (Table 3). In fact, according to Cardoso *et al.* (2003) studies the rate of Rotaviruses was high for less than 12 months old children compared to children who are 24 months old and sharply decreases after 24 months. The frequency of AdE in our sample is 1.9% (12/648) (Table 4). This rate is similar to that obtained by Giordano *et al.* (2001) at Cordoba in Argentina (1.5%), but it is inferior to the rate recorded by Jarecki-Khan *et al.* (1993) in Bangladesh which is 2.8% and that of Fodha *et al.* (2006) in Tunisia (6%). In fact, AdE are mainly acknowledged as important aetiological agents of infant viral gastroenteritis in countries with a temperate climate (Ouermi *et al.*, 2007; Cruz *et al.*, 1990) which could account for the low rate obtained in Burkina Faso, a country of tropical climate.

Parasitology: In addition to the pathological agents showed in evidence, intestinal parasites are also recorded as etiologic agents of AGE: 49/648 *Giardia intestinalis*; 7/648 *Entamoeba histolytica*. Moreover, we could isolate parasites such as *Trichomonas intestinalis* (25/648); *Hymenolepis nana* (16/648); *Strongyloïdae stercoralis* (13/648) that usually do not induce diarrhoea. However, we have not identified Protozoa and Helminths in children under 10 months of life. Among the protozoa identified during our study, *Giardia intestinalis* was the most frequent parasite (7.56%). The giardia are parasites with cysts and their mode of dissemination occurs easily when drinking water or eating dirty vegetables. Nematian *et al.* (2004) have recorded the same facts at Tehran (Iran) with a frequency of 11.5%.

Bacteriology: Among the 648 samples of faeces taken from patients suffering from diarrhoea and analysed in coproculture within the laboratory of Saint Camille, we could identify 267/648 (41.20%) bacterio-positive cultures. At the level of isolated bacteria which cause AGE, the *E. coli EPEC* have the highest frequency (27/72: 37.5%). This prevalence can be compared to other results in Ouagadougou by Bonfiglio *et al.* 2002 (35.0%) which is superior to the one found by Lin *et al.* (2006) (29.7%) by Simpore *et al.* (2008) (27,6%), but inferior to those identified by Abdullah *et al.* (2005) (66.7%) and Mohanna *et al.* (2005) (66.3%). This predominance confirms that the pathogenous *E. coli* remain the major causes of child bacterial diarrhoeas in tropical developing countries (Alikhani *et al.*, 2006; Moyo *et al.*, 2007).

Co-infections HIV: In this study, we emphasize that there is a significant statistic difference between HIV positive and HIV negative as regards co-infections with RV ($p = 0.012$), *Salmonella* sp. ($p<0.001$) and *E. coli EPEC* ($p<0.001$), which means that children that are affected with HIV are the most co-infected with these pathogens because of their immune deficiency. Similarly, children with HIV positive have more AGE and lose more weight than children with HIV negative (WAZ: $p = 0.027$; WHZ: $p = 0.05$).

CONCLUSION

From this study, we notice that: AGE with viral, parasitic or bacterial origin are very important among the pathologies of the less than 60 months old child; many children, irrespective of their HIV serology suffer from diarrhoea and malnutrition and are dehydrated. Any time, HIV seropositive or seronegative children meet high risks of viral, bacterial and parasitic co-infections. Therefore, a control of AGE would need a regular clinical follow up of the children, a veterinary surveillance of the bird and bovine channel as well as an adequate education of young women at procreative age in hygiene, at the levels of Nutritional, Educational and Recuperation Centres (CREN) in Burkina Faso. These types of preventive and training measures could contribute to a significant reduction in the prevalence of AGE, which causes many deaths among children in developing countries.

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