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Prevalence of *Yersinia enterocolitica* among patients in Jos and environs

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Abstract

An investigation on the prevalence and antibiogram of *Yersinia enterocolitica* among patients in Jos and Environs was conducted. A total of 150 stool samples collected from three hospitals namely: Jos University Teaching Hospital (JUTH), Vom Christian Hospital and National Veterinary Research Institute Diagnostic Laboratory, Vom were screened for *Yersinia enterocolitica.* Isolates were identified using standard biochemical tests. 64 (42.6%) of these samples were from male patients while 86 (57.35%) were from female patients. Five (5) *Yersinia entercolitica* isolates were identified. Out of which 4 (6.3%) were recovered from male patients while 1 (1.2%) was recovered from a female. The difference is not statistically significant (P>0.05). Watery and soft stool samples yielded 2 isolates each accounting for the highest isolation among the various forms of stool samples. Age groups 31-40 and 41-50 years yielded two isolates each while one (1) was isolated from the age group 51-60 years. The five (5) isolates were found to be sensitive to gentamicin only. Two isolates were sensitive to chloramphenicol and streptomycin while only one (1) was sensitive to tetracycline.

Keywords: *Yersinia enterocolitica;* Prevalence; Antibiogram; Jos.

Introduction

Yersinia enterocolitica is a Gram-negative rod formerly known as *Bacterium enterocoliticum*. It is now identified as a member of the family Enterobacteriaceae. Studies have shown that the organism is an agent of disease in man. Among the many clinical manifestations of *Yersina* infections in man, the two major ones are acute gastroenteritis usually with blood or non-bloody diarrhoea, abdominal pain and mesenteric lymphadenitis or acute terminal ileitis with symptoms that mimic appendicitis (Sonnenwirth, 1974).

In the last several years *Yersinia enterocolitica* has been isolated from many kinds of clinical and non-clinical specimens in many countries particularly in Europe and Canada (Toma and Lafleur, 1974). Reports from Africa have been scanty. The first report from West Africa was the report incriminating *Yersinia enterocolitica* in a case

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of gastroenteritis and fever in a Nigerian woman (Anjorin *et al.*, 1979)

Since then, others such as Agbonlahor *et al.* (1981; 1983), and Eko and Utsalo (1990) have also isolated the organism. It appears that the general lack of reports from West Africa and Nigeria in particular may be due to either lack of use of proper isolation techniques or that they have not been routinely sought for, from stool samples. The general practice in most clinical laboratories is to culture for the common enteric pathogens without due consideration of other enteric organism like *Y. enterocolitica*. The present study was designed to determine the prevalence of *Yersinia enterocolitica* in Jos and environs and also to determine the antibiogram of the isolates.

Experimental

Samples. The samples analysed in this study included a total of 150 (61 diarrhoeal and 89 non-diarrhoeal) stool specimens collected from patients attending three hospitals in Plateau State. Vom Christian Hospital (63 samples), Diagnostic Laboratory of National Veterinary Research Institute Vom (62 samples) and Jos University Teaching Hospital (JUTH) (25 samples).

Stool specimens were collected from patients in clean, transparent wide-mouthed bottles. Information was also obtained from each subject regarding age, sex, occupation, education, marital status, religion, major symptoms (diarrhoea, vomiting, and fever) duration of the disease, and source of water.

Processing of specimens. The specimens were processed according to guidelines provided by Cheesbrough (1985) on the diagnosis of enteric pathogens. These include:- Macroscopy, cold temperature enrichment, Gram stain, methyl blue test for bipolar staining, motility testing, biochemical testing and antimicrobial sensitivity testing.

Cold temperature enrichment. Stool samples were inoculated into sterile phosphate buffered saline (pH 7.2) and was incubated at 4°C for 3 weeks. The phosphate buffered saline culture was then subcultured into MacConkey agar (oxoid) plates and incubated aerobically at 37°C for 24 hours.

Biochemical testing. Isolates were identified as *Yersinia enterocolitica* using the following standard tests:- catalase test, oxidase test, urease test, citrate utilization test, phenylalanine deamination test, indole test, methyl red test and carbohydrate fermentation test. All tests were as described by Collee and Miles (1989) and Coghlan (1989).

Antimicrobial susceptibility testing. Sensitivity of isolates to antimicrobial agents was determined on Mueller-Hinton agar plates using the disc diffusion method of Scott (1989). From a pure culture of the isolate to be tested, a uniform streak was made on the agar plate. The antibiotic (Antec Diagnostics, UK) discs were placed on the plates and incubated at 37°C overnight. Interpretation of results was done using the zone sizes. Zones of inhibition of $\geq$ 18 mm were considered sensitive, while 12-17 mm intermediate and < 13mm were considered resistant. All isolates were tested for sensitivity to the following antibiotics, ampicillin (10mcg), chloramphenicol (10mcg), cotrimoxazole (25mcg), erythromycin (5mcg), gentamicin (10mcg), penicillin G (5mcg), streptomycin (10mcg) and tetracycline (10mcg).

Statistical Analysis. The data obtained were subjected to the Statview test using a probability of P= 0.05 as the level of significance.

Results and Discussion

Of the 150 faecal samples collected and examined, 64 (42.6%) of these samples were from male patients while 86 (57.3%) were from female patients. 4 (6.35%) of the 64 stool samples from the male patients yielded *Y. enterocolitica* while 1 (1.2%) of the 86 stool samples from female patients yielded *Y. enterocolitica*. The age range of the

patients was from 1-60 years. Of the 150 stool samples examined 5 (3.3%) of the samples yielded *Y. enterocolitica* (Table 1). The highest numbers of isolates were found in two age groups, 31-40 and 41-50 years. Age groups of 1-10, 11-20 and 21-30 years yielded no isolates. The least number of isolate (1) was recovered from the age group, 51-60 years. The difference is not statistically significant (P>0.05).

Table 1: Distribution of *Yersinia enterocolitica* isolated in relation to age and sex

Age group (years)	No of patients sampled		No. (%) positive for *Yersinia enterocolitica*		
	Male	Female	Male	Female	Total
1-10	5	4	0	0	0 (0.0)
11-20	7	13	0	0	0 (0.00
21-30	19	23	0	0	0 (0.0)
31-40	20	23	1	1	2 (5.0)
41-50	10	15	2	0	2 (4.7)
51-60	3	8	1	0	1 (9.0)
Total	64	86	4 (6.3)	1 (1.2)	5 (3.3)
P = 0.2094	P > 0.05				

Table 2: Prevalence of *Y. enterocolitica* according to consistency of stool samples

Consistency of stool samples	Number treated	Number (%) *Yersinia enterocolitica* isolated
Mucoid and blood stained	23	1 (4.3)
Watery	38	2 (5.3)
Soft formed	82	2 (2.4)
Hard formed	7	0 (0.0)
Total	150	5 (3.3)
P = 0.0824,	P>0.05	

Table 3: Other pathogens isolated from patient stool samples.

Pathogens	Number of isolates	Percentage Isolated
Shigella sp.	7	4.67
Salmonella sp.	1	0.6
Total	8	5.27

Table 4: The in-vitro antibiotic susceptibility pattern of the 5 *Yersinia enterocolitica* isolates.

Antibiotic concentration (mcg/mL)	Number of isolates tested	Number (%) sensitive
Ampicillin (10)	5	0 (0.00)
Chloramphenicol (10)	5	2 (40.00)
Cotrimoxazole (25)	5	0 (0.0)
Erythromyin (5)	5	0 (0.0)
Gentamicin (10)	5	5 (100)
Penicillin G (5)	5	0 (0.0)
Streptomycin (10)	5	2 (40.0)
Tetracycline (10)	5	1 (20.00)

Macroscopic examination of the stool samples showed that 82 were soft and 7 were

hard. Watery and soft stool samples yielded 2 *Y. enterocolitica* each while mucoid and blood stained samples yielded 1 (4.3%). Hard stool samples yielded no isolate (Table 2). The difference is not statistically significant (P>0.05). Table 3 shows other organisms causing gastroenteritis isolated from the stool samples. *Shigella spp* topped the list of bacterial isolates in this study, accounting for (53.8) of all the pathogens isolated. *Salmonella spp* was the least organism isolated. An in-vitro antibiotic susceptibility pattern of the 5 isolates of *Yersinia enterocolitica* was determined (Table 4). All the 5 isolates were sensitive to gentamicin. Two were sensitive to chloramphenicol and tetracycline, while none was susceptible to the inhibitory effects of ampicillin, cotrimoxazole, erythromycin and penicillin G.

A total of 150 samples were analysed in this study in which 5 (3.3%) were positive for *Yersinia enterocolitica*. This is high compared with the 1.3% and 1% from diarrhoeal specimens reported by Agbonlahor *et al.* (1981) in Lagos and Eko and Utsalo (1990) in Calabar, Nigeria respectively. This value is also higher than the world wide estimate of 2% reported by Lee (1977).

Transmission of Yersiniosis usually occurs by eating or drinking contaminated food and water or contact with infected animals. Healthy pigs are known to be carriers of pathogenic *Yersinia species* (Wauters *et al.,* 1985). Pig rearing is a common occupation of the indigenous population around Jos and environs and this may be a probable reason for the higher isolation rate of *Yersinia enterocolitica* in this study.

This study showed that 4 (6.4%) of the 64 males and 1 (1.2%) of the 86 females specimens screened were positive for *Yersinia enterocolitica*. The difference is not statistically significance and no sex difference in the distribution of infection has been reported. A probable reason for this finding may be related to the occupation of the two groups, more men would normally be expected to come in contact with infected animals and their products (i.e. during the slaughtering of pigs).

The macroscopic examination of the stool samples indicated that the watery and soft formed stool samples yielded the highest number of isolates 2 (5.3%) and 2 (2.4%) respectively. Although the difference is not statistically significant, this finding agrees with reports in the literature, which showed that diarrhoea caused by *Yersinia* is characterized by watery stools.

In this study, two bacterial genera were also isolated, namely *Salmonella* and *Shigella* species. These bacteria are common etiological agents of gastroenteritis in our environment hence, their presence in diarrheal stool implicate them in the infection.

The five isolates of *Yersinia enterocolitica* were not found to co-infect with the other organisms. This finding indicates that they were probable the primary infecting agents in cases where they were isolated.

The result of in-vitro antibiotic sensitivity test which showed that all the five (5) isolates were sensitive to gentamicin is in agreement with the work of Anyanwu (1985) which showed that all his nine (9) isolates of Yersinia *enterocolitica* were sensitive to gentamicin. Indiscriminate usage of antibiotics (drug abuse) have resulted in multiple drug resistance of many microorganisms in Nigeria (Omonigho *et al.,* 1999). Therefore, the resistance of *Yersinia enterocolitica* isolates in this study to drugs such as ampicillin, cotrimoxazole and penicillin is not surprising. In addition, other enteric bacteria isolated in Jos have also been found to be resistant to these antibiotics (Opajobi *et al.*, 2004).

The present study has shown gentamicin is the most effective drug with 100% success rate. The success of gentamicin

in case management is deemed to be due to its ability to penetrate into joints, tissues and bones. As a broad spectrum antibiotic, gentamicin is effective against most gram negative organisms. Therefore, it could be used as a first line drug for treatment of *Yersinia* infections while awaiting laboratory report. However, gentamicin is administered parenterally which may render usage difficult.

In conclusion this study has shown that even though the isolation rate is low when compared with other enteric pathogens, a small percentage of gastroenteritis can be attributable to *Yersinia enterocolitica* therefore, this bacterium should be sought for routinely in laboratories especially in Jos and environs.

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