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Relationship Between Digestive Tract Pathogens and HIV Infection in Onithsa Metropolis with Respect to Location

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Abstract: Four hundred and seventy eight (478) individuals who exhibited some manifestation of chronic and debilitating illness including persistent cough, skin cancer and dermatitis, multiple lymph adenitis, diarrhea and enteritis, genital sore, urethritis, vaginitis and weight loss were examined to establish relationships between human immuno-deficiency virus infection (HIV) and digestive tract pathogens (DTP) in Onitsha metropolis. There was a significant relationship between HIV positive individuals and digestive tract diseases with HIV positive recording more digestive tract diseases in Onitsha west. Total prevalence of pathogens was 21.63%.

Key words: DTP · HIV and Onitsha

INTRODUCTION

The digestive tract is one of the most common sites for clinical expression of human immune-deficiency virus infection [1, 2, 3 and 4]. Many studies show that victims express oral Candidiasis, approximately one third of patients develop peri-rectal lesions due to Herpes Simplex virus [5, 6, 7 and 8], 30%-80% experience chronic or intermittent diarrhea. Several gastro intestinal diseases appear to be prevalent in these patients including visceral Kaposis sarcoma. microsporidiosis, crytosporidiasis, cytomegalovirus of the digestive tract [9, 10, 11 and 12]. Many HIV infected patients show symptoms of persistent diarrhea, fever and gastritis [13].

The cause of gastroenteritis may be due to proliferation of intestinal opportunistic parasites e.g protozoa, bacteria especially *Salmonella* species, fungi and viruses resulting from lowered immunity [7, 8 and 11]. Persistent gastroenteritis with fever and other symptoms of AIDS including weight loss and dermatological manifestations should raise a warning signal to clinicians and other health workers. It is possible that persistent digestive tract disease may be indicative of HIV infection but the exact relationship between the two is not clear especially with regards to our Nigerian environment which may present different types of intestinal parasites from such types seen in caucatian HIV individuals. It was therere necessary to evaluate intestinal parasites in HIV positive and negative individuals to determine the extent and distribution of the parasites with respect to location.

MATERIALS AND METHODS

Sampled Population: The study examined ur hundred and seventy eight (478) individuals.

Collection of Samples: All inrmation such as location was obtained and data recorded. About 1-2grams of freshly passed stool sample were collected from each patient in a universal specimen container and labeled. While serum samples obtained from coagulated blood samples were used for HIV and Western blot analyses using Savyon Diagnostic Ashdod Israel and Western Blot for confirmatory test by Biorad Novo Blot from Paris, France respectively.

Stool Routine Examination: The stool samples were examined macroscopically for appearance prevalence of blood mucus and texture. A simple normal saline mount was made by picking a little quantity of the stool taken from suspicious sites and mounted in normal saline on a clean glass slide.

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Stool Concentration Technique: All stool samples that yielded no cysts larva, ova or protozoa in the simple normal saline direct wet film examination were subjected to concentration technique as llows:

About 1-2grams of stool were placed in a 20ml glass tubes. About 10ml of 10% normal saline was introduced in each 20ml glass tubes and sieved into small beaker using guaze strainers. This was transferred into 10ml glass tubes *i.e.* 6ml of suspension and 3ml of ether added, mixed well and centrifuged at 3000 rotation per minute for 1 minute.

By means of clean pasture pipettes, a few drops of the deposits were taken and mounted on slides, covered with cover slip and examined under low and high power objective lens of the microscope.

Isolation of Digestive Tract Pathogens from Stool:
Bacteria: Portions of the stool were picked by means of sterile wire loop previously flamed over Bunsen burner and inoculated on Xylose lysine dextrose agar. The inocula were streaked out on the plates using the wire loop flamed in-between streaks to avoid crowded growths in the XLD agar. The plates were incubated at 37°C over night.

Fungus: Using a flamed wire loop, small portions of the stool samples were picked up and inoculated into Sabouraud agar plates, slants and slides in pairs and incubated for 14 days at 37°C also observing daily growths.

Identification of Digestive Tract Bacterial Pathogens: Yellow colonies growing on XLD agar with black centers were suspected Salmonella colonies. These suspected colonies were sub-cultured on Nutrient agar to stabilize the species and the pure growths subjected to biochemical identification as well as motility test by hanging drop method.

Biochemical Identification of Digestive Tract Pathogens: Sterile wire stabs were used to pick the suspected colonies and introduced into Durham fermentation tubes of glucose, lactose, sucrose and manitol. Also, the wire stabs of suspected whoever was pierced through Triple sugar iron agar (T.S.I.A) and sub-cultured onto Mackonkey agar to ensure purity. The fermentation tubes, TSIA tubes and plates were incubated at 37°C over night. All colonies of suspected bacteria that were motile, fermented glucose and manitol with acid and gas, lactose and sucrose not fermented, TSIA slant with alkaline (pink) and acid with gas butt were identified as *Salmonella typhimurium*. The fermentation tubes with motile organisims that fermented glucose, did not ferment lactose and sucrose, acid manitol and orange yellow acid (only) butt of TSIA were identified as *Salmonella typhi.*

Serological Identification of Digestive Tract Bacterial Isolates: A drop of saline was applied on clean glass slide and the bacteria colonies under test on Nutrient agar plates were collected with a sterile wire loop and emulsified on the slide to rm moderate suspensions. Monovalent Salmonella type specific antisera (Oxrd England) were added and steered slightly and observed for characteristic agglutination reaction.

Identification of Digestive Tract Fungal Isolates: The Sabourated agar slants and plates that grew suspected creamy colonies were tested as llows: The colonies were picked up with the tip of sterile capillary pipette and gently transferred and emulsified in 0.5ml sterile serum in a small test tube. Pooled human sera were used.

The above was repeated in a second tube using a known Candida albicans culture as positive control. The tubes were incubated at 37°C for $2\frac{1}{2}$ hours. The sera were mixed and transferred by means of pipette to a slide covered with coverslip. The slides were examined under high power and low powers of the microscope for the presence of short, lateral hyphal filaments (Green tube) rmed by the yeast cells [9].

Identification of Digestive Tract Parasites: The X10 and X40 objectives of the microscopes were used to examine the slides that were mounted with emulsified stools and parasites identified pictorially continuously changing roundish structures with more than ur nuclei identified as *Entamoeba histolytica.*

Roundish and occasionally oblong/oval flagellated highly motile cells were identified as *Trichomonas hominis.* By means of Pasteur pipette, drops of Lugol's iodine were applied to the slides to highlight and confirm the nuclear of *Enta-amoeba histolytica* [9].

Antibiotic Sensitivity Test: Each of the isolates *S. typhimurium* and *S. typhi* were subjected to antibiotic sensitivity as llows; relevant antibiotic mentioned discs e.g *Gentamycin, Amoxycillin, Perflacin, Chloramphenicol, Ampicillin, Tetracyclin, Oflocin,*

Ciproxin, Sporidex, Ciproval and *Ceporex* were placed on each Mueller-Hinton sensitivity solid agar plates that has been previously seeded evenly with the test organisms diluted to $1x10^{4.5}$organisms/ml, disc diffusion method and incubated over night at 37°C.

After incubation, the zones of inhibition surrounding the antibiotics sensitivity discs were taken as a measure of the inhibitory and bactericidal power of the drug against the particular test organisms and denoted as sensitive(s). The discs having the test organisms still growing around the discs were adjudged resistant.

Statistical comparison between digestive tract pathogens in HIV positive and negative individuals were carried out and results noted.

RESULTS

HIV Positive: The thirty-ur cases of Digestive tract diseases yielded eighty two (82) isolates distributed as llows:

Onitsha South recorded highest prevalence of *Trichomonas hominis,* llowed by Onitsha North respectively as llows; 6.67%, 5.50% and next was Onitsha East 4.72%, least was 3.26%. Total = twenty-five (25) *T. hominis.*

Highest prevalence of *Enta-amoeba histolytica* was recorded by Onitsha South 6.0%, llowed by Onitsha West 5.43%, next was Onitsha North 4.59%, least was Onitsha East 1.57%, total twenty-one (21) *E. histolytica.*

Highest prevalence of *Candida albicans* was recorded by Onitsha West 8.70% llowed by Onitsha South, 6.30%, next was 5.33%, least was 1.83%.

Onitsha North recorded highest prevalence of *Salmonella typhi* 1.83%, llowed by Onitsha South 1.33%, next was Onitsha West 1.09%, least was Onitsha East 0.79% total was six (6) *S.typhi*.

Salmonella typhimurium recorded highest prevalence in Onitsha North 1.83%, next was Onitsha South 1.33%, total was ur (4). Onitsha south recorded highest prevalence of total pathogens 21.63%.

HIV Negative: The twenty three (23) cases of Digestive tract diseases yielded thirty nine (39) isolates distribute as seen in the table 1 below.

There was significant difference at P=0.05 in DTP occurrence in HIV positive and negative for locations, Onitsha North and East. There were significant differences however for Onitsha North and East and also for Onitsha South and Onitsha West.

DISCUSSION

Onitsha South recorded the highest number of digestive tract parasites for HIV positives with a prevalence of 6.67% for *Trichomonas hominis*. Other locations, Onitsha North, East and West scored 5.50%, 4.72% and 3.26% respectively. Onitsha South Okpoko, Fegge have very poor hygienic environment with high density population, while Onitsha West is a low density and privileged settlement (GRA). There were generally, more isolates in HIV positives than negatives. Onitsha South (Okpoko and Feege) did not monopolize high prevalence in infection giving impression there was no location specificity as the distribution of isolates were open and even for both HIV positives and negatives. Onitsha South recorded highest prevalence with respect to location for total pathogens, isolated 21.63% for HIV positive individuals. Onitsha South (Okpoko and Fegge) is a low high density area that could record such high figure. Onitsha South and Onitsha West were locations where there was significant difference in occurrence of DTP with more DTP in HIV positive. Other locations showed no significant differences between HIV positive and negative in occurrence of DTP.

Distribution of Digestive Tract Pathogens in Onitsha Metropolis and Their Relations with Hiv Infection with Respect to Location

	HIV Positive						Hiv Negative								
Digestive tract pathogen	ON	OS	OE	OW	Total isolates	Total HIV +ve	ON	OS	OE	OW	Total isolates	Total HIV -ve	Total HIV +ve and -ve	Total HIV +ve and -ve	isolates Total HIV +ve and -ve
Trichomonas hominis (Flagellate)	5.50	6.67	4.72	3	25		2.75	0.96		0.78	1.09	8			
Enta-Amoeba histolytica	4.59	6.0	1.57	5.43	21		0.92	3.33		1.23	0.96	11			
Candida albicans	1.83	6.30	5.33	8.70	26		4.59	3.33		3.9	1.09	16			
Salmonella typhi	1.83	1.33	0.79	1.09	6			1.33		-	-	2			
Salmonella typhimurium	1.83	1.33	-	-	4		-	1.33		-	-	2			
Total 15.38	21.63	12.41		18.48	82	169					39	309		121	478

Onitsha South (Okpoko/Fegge) recorded highest prevalence of digestive tract infection. least was Onitsha East.

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