Relationship Between Digestive Tract Pathogens (DTP) and HIV Infection in Onitsha Metropolis with Respect to Age

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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 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 age group of 20-29 years.

Key words: Urethritis · Vaginitis · HIV And Pathogens

INTRODUCTION

The digestive tract is one of the most common sites for clinical expression of human immune-deficiency virus infection [1, 2]. Many studies show that victims express oral Candidiasis, approximately one third of patients develop peri-rectal lesions due to Herpes Simplex virus [3], 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 [4]. Many HIV infected patients show symptoms of persistent diarrhea, fever and gastritis [5].

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 [6, 7]. 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 was therefore necessary to evaluate intestinal parasites in HIV positive and negative individuals to determine the extent and distribution of the parasites with respect to age.

MATERIALS AND METHODS

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

Collection of Samples: About 1-2grams of freshly passed stool sample were collected from each patient in a universal specimen container and labeled. 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.

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 follows: 1-2grams of stool were placed in a 20ml glass tubes. Also 10ml of 10%

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normal saline was introduced in each 20ml glass tubes and sieved into small beaker using guaze strainers. This was transferred into 10ml glass tubes. 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 [8].

Isolation of Digestive Tract Pathogens from Stool

A.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.

B.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 organisms 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 form moderate suspensions. Monovalent Salmonella type specific antisera (Oxford 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 follows: 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 $2^{1/2}$ 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) formed 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 four 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* [10].

Antibiotic Sensitivity Test: Each of the isolates *S. typhimurium* and *S. typhi* were subjected to antibiotic sensitivity as follows; 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 $1 \times 10^{4.5}$ organisms/ml, disc diffusion method and incubated over night at 37°C [11- 14].

Table 1: Distribution of digestive tract pathogens in onitsha metropolis and their relationship with hiv infection with respect to age

	HIV Positive						Total isolates	Total HIV Positive	HIV Negative						Total isolates	Total HIV Negative	Total HIV Positive and Negative isolates	Total HIV Positive and Negative
	10-19	20-29	30-39	40-49	50-59	60-69			10-19	20-29	30-39	40-49	50-59	60-69				
Trichomonas hominis	6.56	7.56	5.47	3.70	1.89	-	25		1.64	2.52	2.34	0.93	-	-	8			
Enta-Amoeba hiatolytica	3.29	5.58	3.91	2.78	7.55	-	21		4.92	2.52	2.34	1.85	-	-	11			
Candida albicans	3.29	5.58	3.91	7.41	7.55	-	26		8.20	3.36	3.13	2.78	-	-	16			
Salmonella typhi	1.64	0.84	2.34	0.93	-	-	6		1.64	-	0.78	-	-	-	2			
Salmonella typhimurium	-	0.84	1.56	0.93	-	-	4		-	0.84	0.78	-	-	-	2			
Total isolates							82	169							39	309	12	478

There was highest prevalence of digestive tract pathogens in age group of 20-29 years followed by 30-39 years. HIV Positives recorded highest number of isolates of pathogens 82 than HIV negative 39.

RESULTS

HIV Positive: The thirty-four (34) cases of Digestive tract diseases yielded eighty two isolates distributed as follows:

Age group 20-29 years recorded highest prevalence (7.56%) of *Trichomonas hominis* followed by age 10-19 years with 6.56%; next was 30-39 years 5.47%; 40-49 years 3.70% and 60-69 years zero, total was twenty-five (25) *T. hominis.*

Highest prevalence of *Enta-amoeba histolytica* recorded in 50-59 years (7.55%) followed by 20-29 years (5.58%) next was 30-39 years (3.91%) next was 10-19 years (3.29%) least was age group 60-69 years zero, while total was twenty-one (21) *E. histolytica.*

Highest prevalence of *Candida albicans* was recorded in age group 50-59 years (7.55%), followed by 40-49 years (7.41), next was 20-29 years (5.58%), next was 30-39 years (3.91%), next was 10-19 years (3.29%), least was 60-69 years zero, total isolate was twenty-six (26).

Highest prevalence of *Salmonella typhi* was recorded in age group 30-39 years (2.34%), followed by 10-19 (1.64%), next was 0.93%, next was 0.84%.

Statistical analysis of DTP in HIV positive and negative individuals at P= 0.5; df=1 showed there was no significant difference between HIV positive and negative in DTP infection for age groups of 10-19 years and 60-69 years. There were significant differences for other age groups.

DISCUSSION

Age group 20-29 and 10-19 years are generally accepted active age sexually and nutritionally, thus accounting for the high prevalence [4]. However some age groups in HIV negative individuals recorded lower prevalence rates viz 2.52%, for 20-29 years, 2.34% for 30-39 years, 0.93% for 40-49 years, nil for 50-59 years and 60-69 years and 1.64% 10-19 years.

Enta-Amoeba histolytica recorded highest prevalence in age group 50-59 years 7.55%, least was 40-49 years that recorded 2.78%.

There was nothing to suggest that an influencing factor was at play or affecting the isolates from both HIV positives and negative except increase in age. There was decreasing values in prevalence rates generally for both HIV positives and negatives with increase in age.

There was no significant difference statistically in occurrence of digestive tract pathogens in HIV positives and negative individuals (P=0.05) age group 10-19 years and 60-69 years. There was however significant difference in occurrence of DTP in HIV positives and negatives for age groups 20-29 years/, 30-39 years, 40 -49 years, 50-59 years, with more DTP in HIV positives.

Age group 20-29 years recorded the highest prevalence of total pathogens 20.4% for HIV positives. This age bracket represent the youth that is highly exposed to HIV infection including other pathogens.

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