

Epidemiological studies on urinary schistosomiasis and bacterial co-infection in some rural communities of Abia State, Nigeria

Chidinma Ursula Uwandu, Joy Nkeiruka Dike-Ndudim and Chizaram Winners Ndubueze *

Department of Medical Laboratory Science, Faculty of Health Sciences, Imo State University, Owerri, Nigeria.

World Journal of Biology Pharmacy and Health Sciences, 2022, 10(03), 065–072

Publication history: Received on 12 May 2022; revised on 22 June 2022; accepted on 24 June 2022

Article DOI: <https://doi.org/10.30574/wjbphs.2022.10.3.0079>

Abstract

Urinary Schistosomiasis though being second to malaria as the most economically devastating disease is highly neglected in Nigeria. Infant it was only in 2010 that it was introduced into the Integrated Disease Surveillance and Response (IDSR). This has made the control of the disease more challenging despite its known high endemicity in the country. Several studies have shown that the disease can lead some dangerous complications ranging from gross mental retardation in children to renal complications such as bladder cancer. In Abia State Urinary Schistosomiasis is endemic in some rural communities, however not much attempt has been made on bacteria co-infection and to establish some socio – economic variable and clinical presentations carried out a cross – sectional survey of some school children between the ages of 5 – 16 years and above. A questionnaire was administered also. Urine samples were subjected to parasitological and bacteriology studies to determine prevalence, intensity tract infection.

A Urinary Schistosomiasis pooled prevalence of 23.1% in Akwete community, 33.9% in Ozuitem and 0% in Ohuruismiri communities. *Escherichia coli* was the most prevalent micro-organism in co-infection with the prevalence rate of 37.5%, *Klebsiella spp* 15.6%, *Pseudomonas spp* 9.4%, *Streptococcus spp* 6.3% and *Proteus spp* 3.1%.

Keywords: Schistosomiasis; Co-infection; Bacteria; Schistosoma; Urinary Tract Infection

1. Introduction

Schistosomiasis is an acute and chronic parasitic disease caused by blood flukes (Trematode worms) of the genus *Schistosoma* [1]. *Schistosoma* is a cylindrical trematode with separate sexes. They belong to the phylum plathelminthes, class trematode, sub class digenea, super family Schistomatoidea, family Schistosomatidea, genus *Schistosoma* and species *S. haematobium*, *S. mansoni*, *S. Japonicum*, *S. Intercalatum* and *S. mekongi*; they have mouth alimentary canal, but no anus; the adult worms are parasitic in man. All trematode parasites of man and domestic animals are digenetic and endoparasitic.

They require one or more intermediate hosts to complete their life cycles and the name implies they have two generations, sexual and asexual generations [2]. Schistosomiasis is also known as snail fever and biharzia [3]. It is still one of the most deadly neglected tropical diseases and therefore a serious public health problem in Africa, where an estimated 85% of the world's cases are found. Schistosomiasis kills an estimated 280,000 people each year and as the commonest parasitic disease [4] is ranked second to malaria, the disease as with many parasitic diseases is as a result of social inequality and poverty; People get infected because they do not have access to safe potable drinking water and maintain transmission because of lack of proper waste disposal system [5] poor and rural communities (agricultural and fishing population in particular) are more effected by *Schistosomiasis*. People get infected when they perform their

* Corresponding author: Chizaram Winners Ndubueze

Department of Medical Laboratory Science, Faculty of Health Sciences, Imo State University, Owerri, Nigeria.

agricultural duties, get involved in domestic, occupational and recreational activities that bring them in contact with infected water [1]. The infection is reported to have plagued humans since the ancient times [6].

Urinary Schistosomiasis is caused by *Schistosoma haematobium* whose intermediate host is the fresh water snail of the genus *Bulinus* [7]. It is the most prevalent of water-borne diseases, with a very great risk on the health of rural populations [8]. The disease transmission occurs where fresh water snail intermediate host, the genus *Bulinus* is present and where there is contact between the human population and infested water. Subjects who lived close to bodies of water are more exposed and therefore, more vulnerable to *Schistosoma haematobium* infection than those who lived further from the water [9]. Notwithstanding the availability of effective drugs, the annual death rate is around 200,000 in sub-Saharan Africa alone, making this group of parasite the most lethal worm in the world [10]. Over 85% of Schistosomiasis burden is accounted for in Africa and Nigeria is the most endemic country in the world for Urinary Schistosomiasis with an estimated 25.83 million people infected [11, 12]. It was observed to be more prevalent in Nigeria than intestinal helminthes because of the wide urination of egg into water bodies containing the snail host [13],

The public health importance of Schistosomiasis became apparent from its effects on infected individuals and its impact on endemic Communities. The early stages of chronic infection with *S. haematobium* show symptoms and signs which include haematuria, dysuria and pelvic pain and infected person are at the risk of developing renal failure later in life [14]. Many infected persons do not complain, but some can have pain in the bladder regions, the back and occasionally when passing urine. Gross haematuria (visible haematuria), dysuria and increased frequency of micturition [15] are all clinical signs of Urinary Schistosomiasis. Visible haematuria is often transitory but microhaematuria may continue for a long time in an infected person.

Pathological lesion of Schistosome species are caused by the eggs deposited in various tissues [16, 15]. The pass through the tissue aided by the lytic ferments secreted by the miracidium and reach the mucosa of the bladder wall. The presence of the worm results in an endoplebitis making the lumen of the radicals becoming blocked [16]. Papillomata in which numerous eggs occur may develop on the wall of the bladder, the eggs ulcerate through and haemorrhage occurs. Typical diseases manifestation of longstanding infection include anaemia, malnutrition and depending on the infecting *Schistosoma* species, liver, intestines, kidney and bladder diseases develop [17, 18].

Uwaezuoke *et al.*, [19] made an observation of significant bacteria in cases of urinary (vesical) Schistosomiasis and suggests the possibility of secondary bacterial infection with an overall prevalence of 12.6% of significant bacteria in patients with Urinary (Vesical) Schistosomiasis obtained. The prevalence of bacterial infection as a consequence of Urinary (vesical) schistosomiasis has been assessed by other workers. Anosike *et al.*, [20] recorded a prevalence of 67.2% bacteriuria amongst infected persons. Adeyeba and Ojeaga, [21] observed a prevalence of 75.4% bacteriuria in persons infected with *S. haematobium* in their study in Ibadan. The association between Schistosomal and bacterial infections could result from a relationship in which the bacteria either fixed on the cutaneous surface of the worm in clearly defined places [22] or colonize the caecum of the parasite [23]. It is not surprising also that many bacterial species could be isolated in significant amount from Urinary Schistosomiasis patient's urine following the supply of blood to them by the bleeding tissues resulting from the migrating activities of the spined eggs of *S. haematobium*. The torn surfaces bleed, releasing blood for microbial utilization and also provide sites for microbial attachment and proliferation

The distribution of the disease is focal and its effect is more felt in the rural areas of tropics where the population uses natural fresh water habitats for their domestic water supply and or agricultural activities. Hence the disease transmission therefore is contingent on the presence of infested water, the primary snail host are in contact with human population. The severity of *S. haematobium* infection and risk of complications depend in general on the intensity and duration of the infection. Also various studies have been carried out to show the prevalence and intensity of Urinary Schistosomiasis in different parts of Nigeria [24, 25, 26, 27] with little emphasis on the concomitant bacterial infection associated with the disease condition [28, 20]. There seems to be paucity of available reports on the bacteria spectrum and antibiotic activity of the bacteria isolated from such co-infection in Abia State. This study therefore will be timely and of great relevance particularly in the area of intervention.

2. Material and methods

2.1. Study Population

The population for investigation comprised of inhabitants that were selected randomly from the villages of the selected rural communities in Abia State.

2.2. Study Design

This is a prospective case control study involving the parasitological and bacteriological assessment of people having Urinary Schistosomiasis in some rural communities of Abia State. The following Surveys were done:-

Rapid Epidemiological Survey: A survey of the presence of Urinary Schistosomiasis was carried out in the rural communities mentioned.

Malacological Study:- Snail sampling was done using two prominent but parallel methods. To determine snail species and abundance, the first method was the use of metal scoop nets with long metal handles. The second involved the use of rafts which served as snail attractant which is made up of the stems of a creeping plant *Momordica caranta* and prepared according to the method of Opara and Nwoke [29].

2.3. Ethical Consideration

Letter of introduction was collected from the Head of Department (HOD) and given to the Commissioner of Health and that of Health respectively also the community heads of selected areas prior to collection of samples. Informed Consent: Oral written consent was sought from the parents of the subjects before sample collection.

2.4. Selection Criteria

Subjects from the ages of 1 month to 60 and above was randomly selected.

2.5. Exclusion Criteria

While female subjects that on their menstrual cycle were excluded.

2.6. Sample Collection

A urine sample was collected randomly from inhabitants of selected communities. The subjects were instructed on how to collect the early morning urine samples not leaving out the last few drops of urine passed which usually contain the highest number of eggs [30]. This was followed by an oral epidemiological field form.

2.7. Laboratory Processing

The urine specimens were collected between 10.00 and 14.00 hours using wide-mouthed, screw-cap plastic universal containers. The timing was to coincide with peak urinary excretion of Schistosome ova. Urine samples (Midstream Urine) collected from the subjects was analyzed parasitologically and bacteriologically in the laboratory. The Urine sample were collected in 20ml sterile container which was divided into two parts one for parasitological assessment for *S. Haematobium* using a method according to Anosike *et al.*, [20]. The content of the sample container was well shaken after which 10ml of urine was transferred with a sterile disposable syringe to a centrifuge tube and centrifuge for 5min, at 1500rpm. The supernatant was decanted while the sediment was resuspended in the residual urine sample and poured onto a clean dry slide. The sample on the slide was then covered with a clean cover slip and examined under the microscope for the eggs of *Schistosoma haematobium*. Where present the eggs were counted and recorded as eggs/10ml of urine. Haematuria was assayed using chemical reagent strips, Medi-test Combi-9. The test was carried out according to the manufacturer's instructions. The urine specimen were inoculated quantitatively using a calibrated wire loop (that delivers 0.003ml) onto sterile Nutrients agar, Blood agar and MacConkey agar plates for bacteriological examination. The culture plates were incubated aerobically at 37°C for 24 – 28 hours. Samples that show growth of bacterial isolates in a count of $\geq 10^5$ colony – forming units (cfu/ml of urine) after overnight incubation were considered to indicate significant bacteriuria [31]. Characterized isolates were tested for their antibioticogram using commonly used antibiotics.

2.8. Biochemical Tests for Bacterial Identification

The identification of the bacterial isolates were done using biochemical tests as described by Cheesbrough [30]; Cowon and Steel [32]; WHO [7]; Bello *et al.*, [26] and Killic *et al.*, [33]. The following specific biochemical tests will be carried out: Gram staining, Oxidase test, Coagulase test, Indole test, Urease test, Methyl red and voges proskauer test. Analytical Profile index (API) were used for biochemical identification of the bacteria and antibioticogram analysis.

2.9. Antimicrobial Susceptibility Testing

The method described by Cheesbrough [30] were used. The disc diffusion technique; a disc of antimicrobials was placed onto the plate of sensitivity testing agar inoculated with the test organism. The antimicrobial diffuses from the disc into the medium. Following an overnight incubation at 37°C, the culture is examined for areas of no growth around the disc

(zones of inhibition). Bacterial strains sensitive to the antimicrobial were inhabited at a distance from the disc, whereas resistant strains grow up to the edge of the disc.

2.10. Statistical Analysis

The prevalence of infection was calculated using percentages. Tables, proportions and percentages were used to summarize data obtained from the study. The mean of egg intensity was also derived.

3. Results

A total of 142 subjects were studied to isolate Urinary Schistosomiasis and Bacteria co-infection in some communities. Ozuitem community showed the highest prevalence percentage of 33.9% and Akwete 23.1% also Ohuruismiri showed that there is no prevalence from the subjects worked with as shown in table 1. There was more prevalence of Urinary Schistosomiasis in the males than the females in the communities looked at so far. Akwete had 7(24.1%) in males infected which is more than that of the females that is 5(21.7%), Ozuitem had males 13(36.1%) while the females had 7(30.4%) as shown in table 4.2. In table 4.3. 16years and above had the highest prevalence of Urinary Schistosomiasis of 40.0% followed by 13 – 15years 28.3% then 5 – 8 years having 23.5%, with 9 – 12 years having 16.7%.

Table 1 Prevalence and distribution of Urinary Schistosomiasis in some rural communities in Abia State

Communities	No Examined	No Infected	% Infected
Akwete	52	12	23.1%
Ozuitem	59	20	33.9%
Ohuruismiri	31	0	0%
TOTAL	142	32	57.0%

Table 2 Sex related prevalence of Urinary Schistosomiasis in the study area

Communities	Male No Examined	No (%) Infected	Female No Examined	No (%) Infected
Akwete	29	7(24.1%)	23	5(21.7%)
Ozuitem	30	13(36.1%)	23	7(30.4%)
Ohuruismiri	15	0%	16	0%

Table 3 Age related prevalence of Urinary Schistosomiasis in the study area

Age Group (Years)	No Examined	No(%) Infected with Urinary Schistosomiasis	No(%) Infected without Urinary Schistosomiasis
5 – 8 years	17	4(23.5%)	13(76.5%)
9 – 12 years	60	10(16.7)	50(83.3%)
13 – 15 years	60	17(28.3%)	43(71.7%)
16 and above	5	2(40.0%)	3(60.0%)

Table 4 shows that Haematuria is a good indication for Urinary Schistosomiasis in various communities studied so far. In table 5 the prevalence and distribution of proteinuria in the study area was given with Ozuitem giving the highest prevalence of proteinuria with a value of 16(27.1%) followed by Akwete with a prevalence value of 11(21.2%). Table 6 shows the intensity of Urinary Schistosomiasis in the study area analyzed and Ozuitem Community had the highest egg

being produced from subjects with a value of 250 as number of eggs produced and a mean value of 12.5 followed by Akwete community with a value of 130 eggs produced and mean value of 10.8. In table 7 the prevalence of Bacteria involved in co-infection with Urinary Schistosomiasis and *Escherichia coli* being the most prevalent organism with a value of 37.5%, followed by *Klebsiella Spp* 15.6%, *Pseudomonas Spp* 9.4%, *Streptococcus Spp* 6.3% and *Proteous Spp* 3.1%.

Table 4 Prevalence and distribution of haematuria in the study area

Occupational Groups	No Examined	No(%) with Haematuria	No(%) without Haematuria
Akwete	52	10(19.2%)	42(80.8%)
Ozuiem	59	14(23.7%)	45(76.3%)
Ohuruisinmiri	31	0(0%)	31(100%)

Table 5 Prevalence and distribution of proteinuria in the study area

Bacteria Organism	No Examined	No % with Proteinuria	No % without Proteinuria
Akwete	52	11(21.2%)	41(78.8%)
Ozuiem	59	16(27.1%)	43(72.9%)
Ohuruisinmiri	31	0(0%)	31(100%)

Table 6 Intensity of Urinary Schistosomiasis in the study area

Communities	No with Eggs	No of Eggs produced	Mean Egg Output
Akwete	12	130	10.8
Ozuiem	20	250	12.5
Ohuruisinmiri	0	0	0

Table 7 Prevalence of Bacteriuria involved in co-infection with urinary Schistosomiasis

Bacteria Organism	No Examined	No Infected	% Infected
<i>Escherichia coli</i>	32	12	37.5%
<i>Klebsiella spp</i>	32	5	15.6%
<i>Pseudomonas spp</i>	32	3	9.4%
<i>Streptococcus spp</i>	32	2	6.3%
<i>Proteus spp</i>	32	1	3.1%

4. Discussion

This study was carried out to determine the current prevalence of Urinary Schistosomiasis, factors associated with infection and the prevalence of concomitant bacteriuria in Abia State. The socio – demographic data showed that children 13 – 15 years accounted for 42.2% as well as children 9 – 12 years of age 42.2%. This showed higher member of respondents from such age groups might be because of their inquisitive nature and that

children give better compliance. The study also showed a pooled prevalence of 22.2% as against that of Uwazuoke *et al.*, [19]. It is also close to that of Ugbomoiko [9] in three geographical zones in Edo State 22.9%, Okoli and Odabido [11] in selected foci in Imo State (25.1%), Anosike *et al.*, [25] in the Ebonyi Benue River Valley, South Eastern Nigeria (23.5%). A close look at the prevalences of the infection for the communities studied shows variations, which can be interpreted to mean that transmission is highest at Ozuitem (33.9%) followed by Akwete (23.1%). This variation as reported by earlier researchers is attributable to variation in the degree of exposure to infection and presence of bore hole – water in one community than the other. In the present study, the prevalence and severity of infection did not vary so much with sex but did with age, the age 16years and above 40% followed group of 13 – 15 years that had the prevalence of 28.3% followed by 5 – 8 years of age 23.5% and then 9 – 12 years of age 16.7%.

The observation of significant bacteriuria in cases of Urinary Schistosomiasis suggests the possibility of secondary bacterial infection. An overall prevalence of 71.8% of bacteria isolates in subjects of Urinary Schistosomiasis was obtained in this study. The prevalence of bacteria as a consequences of Urinary Schistosomiasis has been assessed by other workers. Anosike *et al.*, [34] recorded a prevalence of 67.2% bacteriuria amongst infected persons, Adeyeba and Ojeoga [21] observed a prevalence of 75.4% bacteriuria in persons infected with *S. haematobium* in their study in Ibadan. The association between Schistosomal and bacteria infections could result from a relationship in which the bacteria either become fixed on the cutaneous surface of the worm in clearly defined places [22] or colonize the caecum of the parasite [23]. It is not surprising also that many bacterial species could be isolated in Schistosomiasis patients urine following the supply of blood to them by the bleeding tissues resulting from the migrating activities of the spined eggs of *S. haematobium*. The torn surfaces bleed, releasing blood for microbial utilization and also provide sites for microbial attachment and proliferation [19].

5. Conclusion

This study has shown that Urinary Schistosomiasis is endemic in Akwete and Ozuitem communities of Abia State, Nigeria and could be a treat to important socio – economic activities in the area. There is urgent need therefore for the Local Government and the State Government to formalize and establish feasible control programmes in the area. This work also suggests that bacteriological examination be carried out for every Urinary Schistosomiasis patient to either confirm or rule out any associated Urinary tract infection. This is to arrest any health implications arising from Urinary tract infections.

Compliance with ethical standards

Acknowledgments

We acknowledge the staff and students of the Department of Medical Laboratory Science, Imo State University, Owerri, Nigeria for their support throughout the period of this work.

Disclosure of conflict of interest

There is no conflict of interest.

Statement of ethical approval

Ethical approval was obtained from the right authority.

Statement of informed consent

Informed consent was obtained from all individual participants included in the study.

References

- [1] World Health Organization. Schistosomiasis Facts Sheet, <http://www.who.int/mediacentre/factsheets/fs/115/en>. 2016.
- [2] Okanula EO, Agba BN, Awotunde JO. "Schistosoma haematobium: Prevalence and Socio-Economic factors among students in cape coast Ghana." African Journal of Biomedical Research. 2003; 6: 69 – 72.
- [3] Colley DG, Bustinday AL, Secor WE, King CH. "Human Schistosomiasis". The Lancet. 2014; 383 (9936): 2253-2264).

- [4] Centers for Disease Control and Prevention. Parasites Schistosomiasis. Available from <http://www.cdc.gov/parasites/schistosomiasis/>. 2014.
- [5] Chitsulo L, Engels D, Montessoro A, Savioli L. "The global status of Schistosomiasis and its control" *Acta Tropica*. 2000; 77: 41 – 51.
- [6] El-Harvery M, Ahmed AMM, Adel Ralman AB, El-Ibiary SA, Ayira AM, Abdel-Hadfez AM, Waheed AA, Hussien H. Schistosomiasis in Egypt: Gambia Governorate". *American Journal of Tropical medicine and Hygiene*. 2001; 62: 42-48.
- [7] World Health Organization. Schistosomiasis progress report (2001 – 2011) Strategic Plan (2012 – 2020) World Health Organization Press, Geneva, Switzerland. 2013.
- [8] Biu AA, Nwosu CO, Akuta A. The incidence of human Schistosomiasis in Maidguri Northern Nigeria. *Bioscience Resource Communications*. 2002; 12(1): 9 – 11.
- [9] Ugbomaiko US, Ofoezie IE, Okoye IC, Heukelback J. Factors associated with urinary schistosomiasis in two peri-urban communities in southwestern Nigeria. *Ann. Trop Med Parasitol*. 2010; 104(5): 409-419.
- [10] Bamgbola FO. Urinary Schistosomiasis *Pediatric Nephrology*. 2014; 29 (11):2113 – 2120.
- [11] Okoli EI, Odabido AB. Urinary Schistosomiasis among school children in Ibadan, an Urban Community in South Western Nigeria. *Tropical Medicine International Health*. 1999; 4(4):308 – 315.
- [12] World Health Organization. Prevention and control of schistosomiasis and soil transmitted helminthiasis. WHO Technical Report series no. 9121 – vi: World Health Organization, Geneva. 2002.
- [13] WHO. Guidelines for the evaluation of soil transmitted helminthiasis and schistosomiasis at community levels. A guide for manager of control programmes WHO/CTD/SIP/98.1. 1998.
- [14] Vander Werf MJ, De Vas SJ, Ngelkerke NJ, Habbema, JD, Engel D. Quantification, of Clinical morbidity associated with schistosome infection in sub-Saharan Africa. *Acta Tropica*. 2003; 86: 125-139.
- [15] Arora DR, Arora B. Trematodes or Flukes. *Medical parasitology (2nd Edition)* B.Sc. Publishers and Distributors. 2009.
- [16] Schmidt D, Roberts L. "Family Schistosomidae: Schistosoma species and Schistosomiasis. *Foundation of Parasitology 7th Edition* McGraw Hill inc. 2006.
- [17] Prata A. Disease in schistosomiasis mansoni in Brazil, Mahmoud A.A.F ed Schistosomiasis London: Imperial college Press. 2001.
- [18] Ouma JH, El-Khoby T, Frenwhich A, Blantan RE. Disease in Schistosomiasis mansoni; in Africa. London: Imperial College Press. 2001.
- [19] Uwaezuoke JC, Anosike JC, Udujih DS, Onyeka PI. Epidemiological and Bacteriological studies on vesical Schistosomiasis in Ikwo Area, Ebonyi State, Nigeria *Journal of Applied Science and Environmental management*. 2008; 12 (2):75-80.
- [20] Anosike JC, Nwoke BEB, Okere AN, Okoro UO, Obiajuru IOC, Ogbulie JN, Ohaeri CN, Ene EO, Njoku CJ. Epidemiological and bacteriological findings in some Schistosomiasis endemic foci in Ebonyi State, Nigeria. *International Journal for Environment Health and Human Development*. 2001b; 2(2):13 -19.
- [21] Adeyaba OA, Ojeaga SGT. Urinary Schistosomiasis and concomitant Urinary tract pathogens among school children in metropolitan Ibadan, Nigeria. *African Journal of Biomedical Research*. 2002; 5:103-107.
- [22] Penand A, Nourrit J, Chapoy P, Alessandrim P, Louchet E, Nicoli RM. Bacteria – Parasite interactions: Enterobacteria and Schistosomes. *Tropical medicine*. 1983; 43: 331-340.
- [23] Ottens H, Dickson G. Studies on the effects of bacteria on experimental Schistosomiasis infections in Animals. *Transactions of the Royal Society of Tropical medicine and Hygiene*. 1972; 66:85 – 107.
- [24] Anosike JC, Okafor FC, Onwuliri COE. Urinary Schistosomiasis in Toro local Government Area of Bauchi State, Nigeria. *Helminthologia*. 1992; 29:77-79.
- [25] Anosike JC, Okere AN, Nwoke BEB, Chukwu JU, Nwosu DC, Ogbusu FI. Endemicity of Vesicle Schistosomiasis in Ebonyi Benue river Valley, Southern Nigeria. *International Journal of Hygiene and environmental health*. 2003; 205 – 206.

- [26] Bello YM, Adamu T, Abubakar U, Muhammad AA. Urinary Schistosomiasis in some villages around the Goronyo Dam, Sokoto State, Nigeria. *The Nigeria Journal of Parasitology*. 2003; 24: 109 -113.
- [27] Nwosu DC, Anosike JC, BEB, Uwaezuoke J. "Epidemiological Assessment of vesical schistosomiasis in Benda Local Government Area of Abia State, Nigeria *Journal of Applied Science and Environmental Management*. 2006; 10(2) :55 - 56
- [28] Alabaraonye FFM, Ekwealor IA. Some aspects of of epidemiology of urinary schistomiasis in Agulu lake, Nigeria. *Journal of Microbiology*. 1995; 10: 9-12.
- [29] Opara AU, Nwoke BEB. Lake Basin area of Anambra State South Eastern Nigeria. The preliminary Report. *Research Journal of Health Science*. 2011; (1):1-5.
- [30] Chessbrough M. *District laboratory practice in Tropical countries*. ECBS Edition. Cambridge University Press. 2002; 280 – 85.
- [31] Gerald LM, Douglas RG, Benette JE. *Principles and Practice of Infectious diseases*. 3rd edition. Churchill Livingstone, New York. 1990
- [32] Cowan ST, Steel KJ. *Manual for the identification of medical Bacteria – 2nd Edition* Cambridge University Press. 2002.
- [33] Killic IH, Ozasian M, Karsligi T, karagoz ID, and Zer Y (2007). Investigation of Diarrhea Agents less than 5 of Age in Summer in Gaziantep/Turkey. *Pak Jour of Bio Sci*. 2007; 10(17): 2915 - 2919.
- [34] Anosike JC, Njoku AJ, Nwoke BEB. Epidemiological and bacterial findings of some Schistosomiasis endermic foci in Ebonyi State, Nigeria. *International Journal of Environmental Health and Human Development*. 2002; 2: 18 – 25.