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THE EFFECT OF *SCHISTOSOMA MANSONI* CERCARIA INFECTION AND TREATMENT WITH NIRIDAZOLE ON TESTICULAR HISTOLOGY OF THE MICE.

BY DAFUR, SYLVNUS JOSIAH* AND LAR, PATRICIA MANKO** *Department of Anatomy, Faculty of Medical Sciences, University of Jos, PMB 2084, Jos, Nigeria. **Department of Microbiology, Faculty of Natural Sciences, University of Jos, PMB 2084, Jos, Nigeria. Correspondent Address: DR. S.J. DAFUR, Department of Anatomy, Faculty of Medical Sciences, University of Jos, PMB 2084, Jos Nigeria.
E-mail: dafurs@unijos.edu.ng, dafurs@yahoo.com

The testicular histology of mice infected with *Schistosoma mansoni* (*S.mansoni*) cercaria and treated with Niridazole was examined. The results reveal that infection of mice with *Schistosoma mansoni* cercariae resulted in distortion of the testicular cyto-architecture including disruption of spermatogenesis as shown by the absence of spermatozoa in the lumen of the seminiferous tubule and destruction of the inter-tubular connective tissue of the infected mice. These changes were reversed to normalcy following two-course treatment of the infected mice with Niridazole after five weeks.

INTRODUCTION

Schistosomiasis of the testis is still rare in many countries. When present, it may simulate cancer resulting in misdiagnosis and invariably leading to orchidectomy, an unfortunate event in the young patient (1). In Africa schistosomiasis of the testis may arise as a result of complication of infestation of the lower urinary tract by *Schistosoma haematobium*. However, a single case of infection of the testis by *Schistosoma mansoni* specie has been reported (2). This shows that both *Schistosoma haematobium* and *Schistosoma mansoni* are capable of affecting the testis, although the mechanism by which this occurs is not clearly understood. It is presumed that in the case of *Schistosoma haematobium* it may be due to retrograde passage of the egg from the urinary bladder into the vas deferens to the testis. The maturation of the *Schistosoma* might occur in the spermatic venous plexus and the deposited egg is carried distally into the smaller vessels of the testis (3).

Although early observations on the pathology of schistosomiasis have been reported (4), only a few reports of testicular schistosomiasis were made in the last sixty years (5,6,7). The pathological changes in this organ following infection are still

incompletely described. Ihekwa reporting a case of testicular schistosomiasis described the pathological changes to include the presence of calcified *Schistosoma* ova, scattered granulomata and fibrous tissue replacement of testicular substance. His report was silent on the state of the seminiferous tubules and spermatogenesis. It is the need to evaluate the effect of testicular schistosomiasis on spermatogenesis and testicular histology that necessitated this study, especially in this era of unexplained male factor infertility, with a view of understanding the mechanisms by which these occur, and hoping to introduce appropriate treatment module.

MATERIALS AND METHODS

i. Source of Parasite Strain

Schistosoma mansoni cercaria was obtained from naturally infected snails (*Biomphalaria pfeifferi*) collected from a stream in Jos, Plateau state of Nigeria where schistosomiasis is endemic. Snails were induced to shed cercaria in beaker containing distilled water by light illumination for one and half hours in the laboratory. The cercaria contained in the distilled water was transferred into a clean beaker and the cercarial density was determined by counting the number of cercaria in 0.5mls or 0.1ml of water.

ii. Source of Mice

Male adult white mice were bought from the animal house of the University of Jos. They were fed with Pfizer mouse cubes and water *ad libitum* and the mice were maintained in cages.

iii. Infection of Mice

The mice were stimulated to defecate and urinate in warm water (25°C) for twenty minutes in a bucket after which they were individually transferred into glass jars which had perforated covers in which about 20mls of distilled water had been placed. In order to infect the mice, between 180-200 cercaria were then transferred into each jar and the mice were allowed to paddle in the water for one and half hours as described by Moore(8).

iv. Treatment of Mice with Niridazole

The infected mice were given 250mls per kg body weight of Niridazole orally daily for five days as one course. After seven days' rest the second course of treatment were repeated. After the first course, 10 mice each from the control and infected groups were sacrificed and the testes obtained and fixed in Buioin's fluid for processing for histology. Five weeks after administering the second course ten mice were sacrificed in each group and the testes also obtained for histology.

v. Histology

the testes were processed for slide preparation and stained with heamatoxylin and eosin examined with the light microscope according to the method of Drury (9).

RESULTS

The control group showed normal testicular cyto-architecture. The seminiferous tubules showed spermatogonial cells at various stages of development and numerous spermatozoa in their lumen. The

interstitial cells and inter tubular connective tissues were intact (Figure 1). The testes of the infected mice with *Schistosoma mansoni* cercaria showed distortion of testicular cyto architecture. The seminiferous tubule revealed disorganized spermatogonial cells and empty seminiferous tubules suggestive of poor spermatogenic activity or disruption in the process of spermatogenesis (Figure 2). There is also destruction of the inter-tubular connective tissue including interstitial cells. This histological picture did not reverse after one course of treatment with Niridazole.

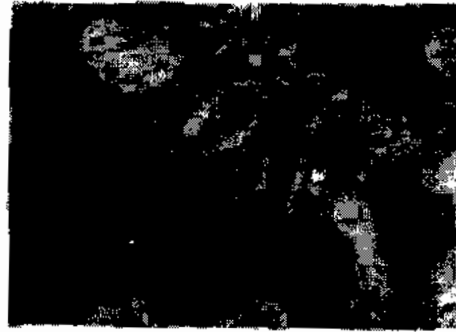


Figure 1: Cross Section of testes of uninfected mice. This shows normal testicular cyto-architecture, testicular cells at various stages of development and numerous spermatozoa in their lumen.

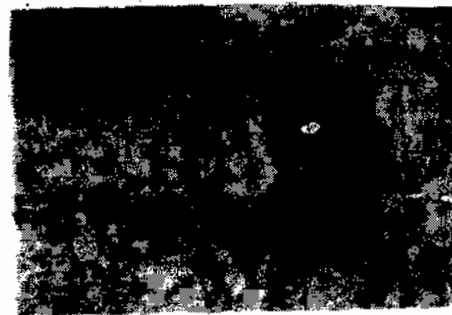


Figure 2: Section of testes of mice infected with cercaria of *S. mansoni*. There is distortion of testicular cyto-architecture. Seminiferous tubules reveal disorganized spermatogonial cells and they are devoid of spermatozoa in their lumen.

Five weeks after treatment of the mice with the second course of Niridazole, the testicular histology showed evidence of return to normalcy. The seminiferous tubules revealed the presence of numerous spermatozoa in the lumen (Figure 3). There is also regeneration of the inter-tubular connecting tissue including the interstitial cells of Leydig. This experimental result reveals that infection of mice with *Schistosoma mansoni* cercaria can result in testicular damage including cessation of spermatogenesis and hence may be a rare cause of male infertility. This damage is however, reversible after five weeks of two courses of treatment with Niridazole.



Figure 3: Section of infected mice testes 5 weeks after completing treatment with Niridazole. Numerous spermatozoa are seen in the lumen of the seminiferous tubules.

DISCUSSION

Our work revealed that infection of mice with *Schistosoma mansoni* induces distortion of the testicular cyto architecture and disruption of spermatogenesis as evidenced by absence of spermatozoa in the lumen of the seminiferous tubules in the *S. mansoni* cercaria treated group. This agrees with the finding of extensive replacement of testicular tissue with fibrous tissue (1). However, we did not find any granulomata or calcified

Schistosoma ova in the testicular tissue. Our finding of poor or scanty spermatozoa in the lumen of the seminiferous tubules is suggestive of disruption in the process of spermatogenesis. In highly endemic area of schistosomiasis such as ours it may be cause of infertility in the male. The mode of spread and mechanisms by which *Schistosoma mansoni* induces these histological change are uncertain. This may be because the incidence of schistosomiasis induced orchitis in the human is rare (2), and therefore, grossly understudied.

Several mechanisms and modes of spread have been proposed for *Schistosoma haematobium* infection, causing testicular pathology. These include retrograde passage of the eggs along the vas deferens to the testes as well as their maturation in the spermatic venous plexus and subsequently carried distally into smaller vessels of the testes (3). In our study no *Schistosoma* ova was seen in the testicular tissue. Given the close relationship between the rectum and the base of the urinary bladder, seminal vesicles, prostate gland and the vas deferens in the male, as well as the rich lymphatic anastomoses around the base of the bladder and rectum; and also the fact that rectal venous plexus which surround the rectum communicates anteriorly with the vesical plexus in the male (10), it is possible for *Schistosoma mansoni* ova whose route to the exterior is the rectum, to enter the vesical plexus and hence the testicular vessels where it may induce immunological response which might result in testicular histo-architectural distortion and disruption of spermatogenesis without the ova being deposited in the testicular tissue.

The finding that these changes were reversible five weeks after treatment with a second course of Niridazole suggest the valuable use of Niridazole in testicular orchitis due to schistosomiasis with or without infertility. It further suggests the need

for a careful investigation of *Schistosoma mansoni* in cases of unexplained male infertility with a view of better management.

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INVESTIGATION OF THE EFFICACY OF TWO RAPID ASSESSMENT TECHNIQUES (OPTIMAL 1 AND SD-BIOLINE) FOR THE DIAGNOSIS OF MALARIA IN RURAL AREAS OF NIGERIA

P.U. AGOMO, V.N. ASIANYA, S.K. AKINDELE, C.O. AGOMO, M.O. AKINYELE, T.A. ADEWOLE, U.T. IGBASI, RC ANYANWU and K.N. EGBUNA. Division of Biochemistry and Nutrition, Nigerian Institute of Medical research, P.M.B. 2013, Yaba, Lagos.

Correspondence: Dr. P.U. AGOMO, Biochemistry Division, NIMR, PMB 2013, Yaba, Lagos

We had previously studied the efficacy of three new techniques-Para Sight ®F, (PSF), Immunochromatographic Test (ICT) and Quantitative Buffy Coat (QBC) – as possible replacements for the time-consuming microscopy in the diagnosis of malaria. Two more rapid assessment techniques (the Optimal 1 and SD-BIOLINE) were recently introduced into Nigeria and claimed to exhibit high sensitivity and specificity. Optimal 1 was particularly claimed to distinguish between *P. falciparum*, *P. malariae*, *P. ovale* and *P. vivax*.

We have in this work evaluated the efficacy of both the Optimal 1 and SD-Bioline in 240 patients from Ibafo and Magboro Communities in Obafemi-Owode LGA of Ogun State, Nigeria. Results showed that with regard to the detection of *P. falciparum*, Optimal 1 gave a sensitivity, specificity, positive and negative predictive values of 63.95%, 92.20%, 82.1% and 82.1% respectively, while the SD-Bioline gave 54.84%, 42.9%, 68.0% and 68.0% respectively. In retrospect, the sensitivities shown by 3 other techniques (ICT, PSF and QBC) investigated by us were 88.63, 89.95 and 87.6% respectively. Their specificities on the other hand were 94.60, 91.17, 94.70% respectively. The main advantage of the rapid Optimal 1 technique is that it was able to detect *P. malariae* which microscopy also detected in three patients. The SD-BIOLINE gave the worst comparative result and could not be recommended for use in Nigeria. This work in conclusion has shown that Optimal 1 could be useful in the rapid diagnosis of the various species of *Plasmodium* in Nigeria provided the patients could afford the test.

Key Words: Malaria, Optimal 1, SD-Bioline, *P. falciparum*.

INTRODUCTION:

Since its introduction by Ross in 1903, the thick blood film microscopy (TFM) has been the primary method of malaria diagnosis throughout the world (1). Despite the simple technology, this technique is still expensive and requires adequate infrastructure to maintain, train health workers and ensure quality assurance of the service. Microscopy also requires electricity, which is not available in most Health centres in rural areas. As a result of these limitations, laboratory support which is needed due to problems of over-prescription of antimalarial drugs and drug-resistance is not available in Primary Health care centres where clinical diagnosis and presumptive treatment are the only method adopted. The urgent need for a rapid, simple, reliable and cost-effective diagnostic technique to overcome the deficiencies of light microscopy has been recognised for a long time. In 1993, rapid diagnosis of malaria was made a key feature of the world Health Organisation Global Malaria Control Strategy so that effective treatment could be administered quickly to reduce morbidity and

mortality especially in children and pregnant women (2,3). Rapid diagnosis is also a key feature of one of the six elements of the Roll Back Malaria Programme (4).

Some scientists have made attempts at developing alternative techniques (5). Some of the early attempts concentrated on the improvement of the sensitivity of light microscopy by concentrating the parasites in the blood sample, improving the visualization and detection, staining with fluorescent dyes or, a combination of these approaches such as the quantitative buffy coat technique (QBCT) (6) and the kawamoto fluorescent technique (7). Newer methods which have been claimed to be better than the TFM were based on the identification of parasite-specific antigens and molecules such as nucleic acids, the sensitivity in some cases being improved by the use of the polymerase chain reaction (PCR) technique (5). All these techniques utilize sophisticated equipment and electricity, and thereby suffer from similar logistic problems as the microscopic technique. A new direction was introduced when Howard et al described the secretion of a malarial histidine-rich protein-2 from

Plasmodium falciparum infected erythrocytes(8). A rapid technique, ParaSight®F was then developed by Becton Dickinson Tropical Diseases Diagnostics, Sparks, MD. This was based on the detection of *P. falciparum* histidine-rich protein-2 in human blood within 10 minutes and without the use of electricity. The usefulness of ParaSight®F was reported in Thailand(9) Tanzania(10) Brazil(11) Zimbabwe(12) and Kenya(13). In our laboratories, preliminary evaluation of ParaSight®F in 235 patients in Nigeria expressed some reservation considering the cost of the strips and the spill-over effect of antigenaemia long after parasitaemia had cleared(14). However, further work with a larger number of patients (501) presented the benefits of ParaSight®F in better light especially when the results were stratified according to the various health facilities and Local Governments (15). A new technique, the Immunochromatographic Test (ICT) was introduced not long after the introduction of ParaSight®F. Although it is based on the same principle of detection of the histidine-rich protein-2 in *P. falciparum* infection, the manufacturers (ICT Diagnostics, Sydney Australia) and some investigators outside Nigeria claimed that it is an improvement on all other rapid assessment techniques (16,17,18,19). The ICT has been evaluated in our Laboratory (20) and its comparative assessment with ParaSight®F in two Local Government Areas showed the former to have an edge over the latter, although no statistical conclusions could be drawn(21). Another addition to rapid antigen detection tests is the optimal 1 assay (Flow Inc) which has been claimed to detect all four species of malaria parasites, namely, *P. vivax*, *P. falciparum*, *P. malariae* and *P. ovale*(22). The optimal 1 technique is

said to detect the metabolic enzyme, parasite lactate dehydrogenase (pLDH) produced by viable malaria parasites and also released from parasite-infected erythrocytes. Differentiation of the malaria species is said to be based on the antigenic differences between the pLDH isoforms. The presence of pLDH in the blood is also claimed to reflect the presence of viable malaria parasites. Optimal 1 is new in Nigeria and we are not aware of anyone who has undertaken the evaluation of the efficacy of this technique in Nigeria. There is however evidence of its review by Bailey(11) and its evaluation in Honduras(22). We therefore deemed it useful to investigate the performance of Optimal 1 in order to compare its efficacy with those of the microscopic technique and other rapid assessment methods for possible utilization in rural areas.

Also new in Nigeria is another rapid assessment device, SD-BIOLINE, for the diagnosis of *Plasmodium falciparum*. The SD BIOLINE *Plasmodium falciparum* test is claimed to be rapid test for the qualitative detection of antibodies of all isotypes (IgG, IgM, IgA) specific for *Plasmodium falciparum* in human serum, plasma or whole blood. The SD BIOLINE *Plasmodium falciparum* test is said to contain a membrane strip which is pre-coated with recombinant *P. falciparum* capture antigens, merozoite surface protein (MSP) and Circumsporozoite protein, (CSP) on the test band region. The principle of this device has it that the recombinant malaria *Plasmodium falciparum* antigen (MSP & CSP) - colloid gold conjugate and blood/serum sample move along the membrane chromatographically to the test region and form a visible line as the antigen-antibody gold particle with a high degree of sensitivity and specificity. In a comparative analysis of the SD BIOLINE *P. falciparum* test versus microscopic examination of whole blood, the manufacturers recorded a sensitivity of 88.2%, a specificity of 98.5% and a total agreement of 93.8%. These claims

prompted us to investigate the performance of the two new techniques in Nigeria in comparison with other rapid assessment techniques.

MATERIALS AND METHODS

Study sites and Study subjects:

Ibafo-Magboro group of communities is the rural parts of Obafemi-Owode LGA, Ogun State lies 80 KM east of Lagos. Most of the 18 villages in the area have neither electricity nor pipe borne water. The only Police Station is in Ibafo town which is the only developed part of the group of Communities. Their LGA headquarters is in Owode near Abeokuta, about 50km away. Most of the villages are surrounded by thick forest although in recent times they have started selling the wooded lands to some organizations which are now cutting down some of the trees. A total of 240 study subjects (65 adults and 175 children, 133 females and 107 males) were randomly recruited for this work from some of these villages after obtaining consent from heads of the communities and from the individuals or their guardians. The patients recruited complained of fever, aches, pains, loss of appetite, bitter taste and other malaria-associated symptomatology. Some patients with non-specific complaints were also part of the subjects included in the project. Following an explanation and consent by the patient, a simple questionnaire was completed as regards age, sex, current medication, any recent blood transfusion and malaria associated signs and symptoms.

Microscopic Examination

The technique used for microscopic identification was as reported in our previous work^{14,15,21}. The presence of malaria parasites using this microscopic technique was

considered as the "Gold Standard" reference level for positively. Parasite enumeration and identification of the species were carried out by two, and in some cases, three microscopists for every specimen collected. This was for purposes of accuracy and confirmation of results.

The Packed Cell Volume (PCV) of each patient was determined as in previous work, using capillary tubes and haematocrit centrifuge. The results were read with a haematocrit reader.

Optimal 1 was supplied as the Diamed Optimal Test Kit and used as instructed by the Manufacturers. The principle has been enunciated (see introduction).

Briefly, in the procedure, a dipstick was introduced to lysed blood and this was followed by a buffer (supplied). After 12 minutes the strip was examined. A negative result showed on a reaction line, those of *P. ovale* (supplied) and *P. malariae* showed two positive lines while the presence of *P. falciparum* indicated 3 lines. Only viable parasites were said to produce lactate dehydrogenase.

The SD BIOLINE Malaria P.falciparum test device:

The Principle of this test has been explained in the introductory part of this paper. The test device was removed from the foil and placed on a flat, dry surface. Whole blood (20ul) was slowly added to the sample well and this was followed by 3 drops of the assay diluent. As the test began to work a purple colour moved across the result window in the centre of the test device. The result was interpreted within 5-20 minutes, but not after 20 minutes. The colour band appearing in the left section of the result window was the control and it indicated that the test was working properly. The presence of only one band within the result window indicated a negative result whilst the presence of two bands

within the result window, (regardless of which band appeared first) indicated a positive result.

Studies on the Disappearance of parasitaemia, Lactate dehydrogenase (LDH) and BIOLINE antibodies.

A Cohort Study was also undertaken to examine the test performance over a period of time in 10 patients known to be parasitaemic. Following positive diagnosis, patients were treated with therapeutic doses of Fansidar® or Chloroquine. Venous blood was subsequently collected on day 4 and day 7 by finger prick in order to follow the decline in parasitaemia and antigenaemia. This test was only carried out in ten (10) positive patients since it could not be done in all the patients as a result of limitation in the number of test strips.

Analysis of Results: Data were entered in the EPI-INFO 6.04 and analysed. Values were given in the text and tables as means ± SD where applicable. Values for different groups (age, etc) were compared using students t-test or chi squared test and P. values less than 0.05 were taken as significant. analysis of variance was performed for multiple group comparisons. The key variables were as follows:

$$\text{Sensitivity (\%)} = \frac{\text{No of True Positives (TP)} \times 100}{\text{No of TP} + \text{No. of false Neg(FN)}}$$

$$\text{Specificity (\%)} = \frac{\text{No of True Negatives (TN)} \times 100}{\text{No of TN} + \text{No of false Positives (FP)}}$$

$$\text{Positive Predictive Value (PPV)\%} = \frac{\text{No of TP} \times 100}{\text{No TP} + \text{FP}}$$

$$\text{Negative Predictive value (NPV)} = \frac{\text{FN} \times 100}{\text{FN} + \text{TN}}$$

$$\text{False Negative Rates (FNR)\%} = \frac{\text{FN} \times 100}{\text{FN} + \text{TN}}$$

RESULTS

COMPARISON OF MALARIA PREVALENCE RATES USING THICK/THIN BLOOD FILM MICROSCOPY, OPTIMAL 1 AND SD-BIOLINE TECHNIQUES.

Data showed that of a total of 240 subjects examined by microscopy and the Diamed Optimal (Optimal1), 86(35.8%) were Parasitaemic by microscopy while 67 (27.9%) were positive with Optimal 1 rapid assessment technique (Table I). Three (3) patients had P. Malariae which were identified by both techniques. With the removal of the P. Malariae, prevalence by microscopy was 34.6% while that of optimal 1 was 26.7%. mixed infections (3 in number) were also identified by both techniques.

The group investigating the performance of SD-Bioline undertook the test in 45 patients most of who were clinically malaria-positive. Prevalence rate of 55.6% using the BIOLINE and 68.9% using microscopy were obtained but these could not in fact be taken as true prevalence rates of parasitaemia in the area. Random investigation was considered a better technique for determination of prevalence.

Spill-Over effect of Antigenaemia:

Re-investigation of parasitaemia by light microscopy after anti-malarial therapy showed that the 10 randomly-selected previously-infected patients were negative on day 4 and on day 7 when they were re-examined. Conversely on that 4th day, 2 (20.0%) and 3(30.0%) of the patients were still Optimal 1 and SD-BIOLINE positive respectively. Further test for antigenaemia using these rapid assessment techniques were not undertaken on day 7 and 14 as a result of the limited number of the test strips available. However, negative microscopy on day 4 for all the previously positive patients indicated that there was no antimalarial resistance in the patients. The positive

result obtained with the test strips after 4 days of chemotherapy in a number of patients mentioned above could only indicate the persistence of antibodies long after parasitaemia must have been cleared from the blood.

Table I: Sensitivity and Specificity of the DiaMed Optimal for the detection of *P. falciparum* in 240 patients in Ibafo-Magboro Communities of Obafemi-Owode LGA, Ogun State.

THICK BLOOD FILM MICROSCOPY				
DIAMED OPTIMAL I		Positive	Negative	Total
	Positive	55	12	67
	Negative	31	142	173
	Total	86	154	240

$$\text{Sensitivity: } \frac{55}{86} \times 100 = 63.95$$

$$\text{Specificity: } \frac{142}{154} \times 100 = 92.20$$

$$\text{Positive Predictive Value: } \frac{55}{67} \times 100 = 82.1$$

$$\text{Negative Predictive Value: } \frac{142}{173} = 82.1$$

Table II: Sensitivity and Specificity of SD-BIOLINE for the detection of *P. falciparum* in-patients in Ibafo Communities of Obafemi-Owode LGA, Ogun State.

THICK BLOOD FILM MICROSCOPY				
SD-BIOLINE		Positive	Negative	Total
	Positive	17	8	25
	Negative	14	6	20
	Total	31	14	45

$$\text{Sensitivity: } \frac{17}{31} \times \frac{100}{1} = 54.84\%$$

$$\text{Specificity: } \frac{6}{14} \times \frac{100}{1} = 42.9\%$$

$$\text{Positive Predictive Value: } \frac{17}{31} \times \frac{100}{1} = 68.0\%$$

$$\text{Negative Predictive Value: } \frac{6}{20} \times \frac{100}{1} = 68.0\%$$

SENSITIVITIES AND SPECIFICITIES OF THE DIAMED OPTIMAL AND SD-BIOLINE USING MICROSCOPY AS THE GOLD STANDARD

Result using the DiaMed Optimal gave a sensitivity, specificity, positive and negative predictive Values of 63.95%, 92.20%, 82.1% and 82.1% respectively. The SD-BIOLINE gave 54.84%, 42.9%, 68.0% and 68.0% respectively. In retrospect, the sensitivities shown by 3 other techniques (ICT, PSF and QBC) investigated by us were 94.60%, 91.17%, and 94.17% respectively (Table I). The SD-BIOLINE gave the worst comparative results and could not be recommended for use in Nigeria

DISCUSSION

We have investigated the efficacy of two rapid techniques for the diagnosis of *P. falciparum* in 240 patients from Ibafo and Magboro Communities, which are rural areas of Ogun State. in this context, we have shown that the SD-BIOLINE was not able to detect some

positive cases which Optimal 1 (DiaMed Optimal) and microscopy could detect. However one interesting aspect of SD-Bioline is that the manufacturers used two surface proteins, namely, merozoite surface protein (MSP-1) and circumsporozoite protein (CSP) which have previously been documented to induce potent antibodies. They have been characterized and have been put forward as strong candidates for malaria vaccine development (23,24,25). It would have been interesting if this rapid assessment dipstick (BIOLINE) had surpassed all others in the detection of *Plasmodium falciparum* since it would have been seen as playing a dual role in our quest for methods of controlling malaria. Nevertheless, we are still investigating why the sensitivity and specificity of SD-BIOLINE were as low as 54.84% and 42.9% respectively while those of Optimal 1 now marketed as DiaMed Optimal by some companies were 63.95% and 92.20% respectively. The expiry date of the former was unfortunately not boldly printed on the packet. Comparative results using our previous work in 12 sites (manuscript accepted in journal of Malaria in Africa) has not shown the present work with Optimal 1 to be the best rapid assessment technique for malaria diagnosis. The immunochromatographic Test (ICT), ParaSight® F (PSF) and Quantitative Buffy Coat (QBC) techniques gave mean sensitivities as 88.63%, 89.95% and 87.6% respectively. The mean specificities were 94.60%, 91.17% and 91.7% respectively. These, especially ICT, seemed to be better than Optimal 1 although no statistical significance emerged. One advantage of Optimal 1 is its ability to detect and distinguish between the various isoforms of the metabolic enzyme, Lactate dehydrogenase (pLDH) of the various

species of malaria parasites. However the work using the DiaMed Optimal (Optimal 1) and SD-BIOLINE was done in just one section of Obafemi - Owode LGA. It is possible that when more kits become available and a larger number of patients are tested in various communities and sites as in PSF and ICT, the main benefits, especially of Optimal 1 will emerge.

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Evaluation of the OptiMAL Test for Rapid Diagnosis of Malaria

¹UJAH I.O.A., ²IKEH, E.I., ³GLEW RH, ³VANDERJAGT, D.J. Departments of Obstetrics and Gynaecology¹ and Medical Microbiology² Faculty of Medical Sciences, University of Jos, Nigeria and Department of Biochemistry & Molecular Biology, University of New Mexico, Albuquerque³

Correspondent Address: Dr. E.I. IKEH, Department of Medical Microbiology, Faculty of Medical Sciences, University of Jos P.M.B. 2084, Jos.

This study evaluated the ability of a newly developed rapid test for laboratory diagnosis of malaria. OptiMAL is a rapid test that utilizes a dipstick coated with monoclonal antibodies against the intracellular parasite dehydrogenase (PLDH). The differentiation of Plasmodium species is based on antigenic differences between the PLDH forms.

Blood samples from 62 of clinically diagnosed patients were examined using the microscopy of Giemsa-stained blood films and the OptiMAL test. The blood films indicated that 27% of the patients were positive for *P. falciparum* (including one case of mixed infection with *P. malariae*), while the OptiMAL test recorded 34% for *P. falciparum*.

The OptiMAL test failed to diagnose malaria at concentrations less than 100 per microliter of blood, while those missed by microscopy may be due to sequestration of the parasite coupled with low parasite density. The OptiMAL test was modified by using fingerprick instead of venepuncture and this simplifies the test both in terms of cost and trained personnel. There was no significant difference between the two methods ($\chi^2 = 1.513; P > 0.05$), but the

OptiMAL test has the advantages of being faster, requires almost no specialized laboratory experience and extremely sensitive and specific even in field situations. We conclude that the OptiMAL test is an effective tool for the rapid diagnosis of malaria.

INTRODUCTION.

Malaria has had resurgence in many tropical countries. The disease now occurs in more than 90 countries worldwide, and it is estimated that there are over 500 million clinical cases and 2.7 million malaria-caused deaths per year (1). Although tremendous efforts and resources have been invested in the study, control and probably eradication of malaria, it continues to be a major problem particularly in the rural areas of tropical Africa. In Nigeria, malaria due to *Plasmodium falciparum* is holoendemic in many parts of the country (2,3). Multitudes of factors have contributed to the continued persistence of malaria. Some of these are: (i) insecticide resistance in the *Anopheles* mosquito. (ii) Social instability resulting in movement of unexpected non-immune individuals into the endemic areas and (iii) Failure to develop an effective vaccine (4). Compounding the problem of increasing morbidity and mortality are the emergence and rapid spread of anti malaria drug resistance which necessitate the use of more expensive and sometimes toxic anti malaria and longer treatment course (5).

The ability to diagnosis malaria early and the prompt treatment of infected individuals are very fundamental in the prevention and control of malaria.

Clinical diagnosis of malaria can be very imprecise and should be confirmed with laboratory diagnosis. Definitive diagnosis is currently based on microscopic examination of stained thick and thin blood films and malaria control activities therefore require laboratories. Thus diagnosis of malaria by this method is time consuming and requires considerable expertise for its interpretation.

Recently, a new malaria detection test, OptiMAL (Flow Inc., Portland, Oregon) was introduced. This test is based on detection of parasite lactate dehydrogenase enzymes (PLDH). The aim of this study was to evaluate the diagnostic accuracy of the new optiMAL test as compared to the traditional method.

MATERIALS AND METHODS

Study Area: The location, topography, climate and other geographical indices of Jos where the study was conducted are given somewhere else (6).

Study Population: The population studied consisted of 62 infants and adults resident in Jos with symptoms suggestive of malaria. Informed consent was obtained from the patients or their relations before being recruited in the study and the Helsinki revised declaration principle was strictly adhered to.

PARASITOLOGICAL TECHNIQUES

(a) Collection of Samples:

Two milliliters of venous blood was collected from the patients and thin and thick blood films were made on the same slide. The remaining sample was placed in the EDTA bottle and mixed gently. The anticoagulated samples were refrigerated at 4°C if not immediately required for the OptiMAL test. Finger - pricking method was used for the sample collection in a few of the patients, especially infants.

(b) Malaria Diagnosis with thin and Thick Blood Films.

The thin and thick blood films were stained appropriately using the Giemsa's staining method and analysed microscopically at X 1000 magnification for the presence of malaria parasites. The average time spent per slide varied depending on the number of parasites present in the blood sample. The study was blinded, since results of microscopy were not shared with the individual performing the optiMAL test until all samples were processed. Parasitemia levels were calculated with results from stained thick films and parasites were counted in 100 consecutive fields.

Parasite densities were calculated by assuming 0.2 microliter of blood per thick film and that each microliter of blood contained 8, 000 leucocytes (7,8).

(c) Malaria Diagnosis with OptiMAL.

All the samples were tested with the OptiMAL assay. Briefly 0.05 millilitre (2 drops) of blood was added to 2 drops of reagent A. and mixed. The sample was then allowed to migrate to the top of the OptiMAL dipstick. After 8 minutes, the optiMAL strip was cleared by adding 4 drops of reagent B. The appearance of 3 dark bands on the strip indicates *P. falciparum* infection while 2 dark bands indicates either *P. malariae*, *P. vivax* or *P. ovale* infections. A positive control band appears at the top of each strip as an indicator that the test is working correctly. Statistical analysis was done using the chi-square at 5 percent level of significance.

RESULTS

Of the 62 patients sampled whose clinical conditions suggested malaria infection, 22 (35.5%) were positive for malaria. The blood film results indicated that 27% (17 of 62) of the patients were infected with malaria based on the morphologies of the various stages of the parasite. Among the patients' *P. falciparum* was present in all the positive blood films while there was only one mixed infection of *P. falciparum* and *P. malariae*. Correspondingly, the optiMAL test results indicated that 34% (21 of 62) of the patients' samples were positive for malaria parasite. The mixed infection of *P. falciparum* and *P. malariae* was not differentiated with the optiMAL test and in this particular case showed three bands (see Table 1).

TABLE 1: MALARIA PARASITE DETECTION USING OptiMAL TEST AND GIEMSA - STAINED BLOOD FILMS

Species	OptiMAL Result		Blood Film Result		Total
	Positive No %	Negative No %	Positive No %	Negative No %	
<i>Plasmodium falciparum</i>	21 (34)	41 (66)	17 (27)	45 (73)	62
<i>P. malariae</i>	1 (100)	NA NA	1 (100)	NA NA	1

The blood films indicated one *P. falciparum* positive sample that was not identified by the OptiMAL test. The parasite density in this case was not identified by the OptiMAL test. The parasite density in this case was 80 per micro litre of blood. However, OptiMAL test detected five cases of *P. falciparum* infection that were not detected with the stained blood films. One of these patients had already been treated with chloroquine phosphate before the OptiMAL test was carried out. The parasite density ranges from 80 to 7,120 per micro litre of blood. The trophozoites and gametocytes of *P. falciparum* were present in the positive samples while the trophozoites and schizonts of *P. malariae* were found in the one case of mixed infection.

DISCUSSION

The study compared the diagnosis of malaria by a new rapid test (OptiMAL) with the traditional microscopy of stained films and found that the two methods yielded comparable results as there was no significant difference between the two methods ($X^2 = 1.513$; $P > 0.05$). The one case of malaria that was missed by the OptiMAL test may be due to the insensitivity to low parasitemia levels which in this case was 80 per microlitre of blood. It could also be attributed to the fact that the OptiMAL test detects only viable parasites. The one case where the OptiMAL test was negative and the stained blood films detected the parasite was because the patient had already been treated with chloroquine phosphate before the sample was collected. There were 5 cases in which the OptiMAL test was positive while the stained films were negative.

This may be due to the sequestration of the malaria parasites. Although the difference is not statistically significant, it shows that even with a very good microscopist, there is the possibility of missing a few of the malaria cases. It was also observed that the OptiMAL test was positive even after the anticoagulated sample had been stored at 4°C for 7 days. In the case of the blood films, the parasitemia levels decreased on storage, until the stained films became negative on the third day. This makes it difficult to diagnose malaria at a more convenient time when the materials would be available.

There are compelling reasons to justify the use of a rapid malaria diagnostic test, especially in developing countries. Definitive diagnosis of infection in malaria control is currently based on microscopy and malaria control activities must therefore require the use of laboratories. The advantages of the OptiMAL test are that (a) it is fast (b) it requires almost no additional laboratory experience, (c) it is extremely sensitive and can be carried out in field situations. This, we believe is an invaluable malaria diagnostic tool of the future. It would do away with the need for slides, their transportation to centralized laboratories and the required infrastructure and trained staff and in situations where the electricity supply is very erratic like Nigeria, the OptiMAL test is a perfectly suitable alternative. This will certainly improve the diagnostic accuracy of malaria at the primary health care level and by community health workers.

The only drawback of the OptiMAL test is that the cost per unit

diagnosis may be currently high and its present use in malaria control programmes may only be cost effective in areas of low transmission where the cost of setting up a microscopic laboratory and equipment exceeds the cost of OptiMAL diagnosis. The OptiMAL test can be modified by using finger-prick samples instead of the venepuncture. This further simplifies the test both in terms of cost and trained personnel and therefore this method of collecting blood sample for the OptiMAL test is advocated.

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MALARIA MORBIDITY AMONGST HOSPITAL WORKERS IN ILORIN

ADEMOLA O. AWOYEMI

Department of Epidemiology and Community Health
University of Ilorin, Ilorin Nigeria.

A study of malaria as a cause of morbidity among the staff of the University of Ilorin Teaching Hospital, Ilorin, Nigeria was carried out from August to October 2001. Patients attending a community based Health Centre was used as control. Malaria accounted for 43.0% of illnesses among the workers and 36.7% in the general public. Malaria was responsible for nearly two-thirds of sickness absence and for 48.6% of days lost due to illnesses. Also the disease was responsible for 50 percent or more of sickness absence among all categories of workers when analyzed by occupations. These findings show that malaria could disrupt the health care delivery of a country since it could affect even health workers. By extension too, it could result in the disruption of the economic activities of the country and result in low productivity if not effectively controlled. It therefore recommended that all efforts at effective controlling malaria should be put in place in Nigeria.

INTRODUCTION

Three to five hundred million cases of malaria occur annually in the world causing over one to two million deaths especially among children (1). The disease typically presents with fever and diagnosis is based on microscopic demonstration of malaria parasites in the blood of victims (1,2). While malaria is known to be endemic in the tropics and sub-tropics, its impact on the working class has only been scantily documented in the literature (3).

The hospital workers especially are expected to know about preventive measures against malaria - hence on this assumption are less likely to contract the disease compared with the general public. In addition, not much information is available in the literature about morbidity pattern among hospital workers (4,5).

It is in the light of this dearth of information that this study was undertaken in order to determine the extent of morbidity from malaria among workers of the University of Ilorin Teaching Hospital, Ilorin, Nigeria attending the Staff Clinic of the Hospital over a period of three months.

MATERIALS AND METHOD

The study was undertaken from 1st August to 31st of October, 2001 in Ilorin metropolis. It involved a study group, the staff of the University of Ilorin Teaching Hospital, Ilorin, Nigeria attending the Staff Clinic of the Hospital within the period. The control group was made up of patients attending the Okelele Health Centre from Okelele community of Ilorin.

The year 2000 mid-year working population of the Teaching Hospital was 2,058 according to the Hospital Annual Report (12). In the same year the bed complement was 486. The staff clinic was run by 5 Consultants, 6 Resident doctors, 6 Community Health Nursing Officers and 9 Community Extension Workers during the period of study.

The Okelele Community Health Clinic which serves the Gambari ward community of Ilorin is run by doctors and nurses from the same Department of Epidemiology and Community Health which also supervises the Staff Clinic of the Teaching Hospital. Patients are seen here and treated by doctors and other health workers while difficult cases are referred to the Teaching Hospital.

The study was carried out concurrently in the two identified populations. In the study group, patients who reported at the Staff Clinic with fever and symptoms suspected to

be due to malaria were sent to the Haematology Laboratory for confirmatory evidence of malaria parasites. Only those so confirmed were included in the study.

Among the control group, blood samples were also obtained at the Okelele Health Clinic in patients who reported with fever. The samples were also sent to the Haematology Laboratory of the Teaching Hospital for confirmation of diagnosis in order to standardize procedures. Other diseases diagnosed and treated at the two clinics during the period were also recorded.

RESULTS

During the study period—August to October, 2001, a total of 1963 medical conditions were treated in the Staff Clinic of the University of Ilorin Teaching Hospital, Ilorin while 1397 medical conditions were seen at the Okelele Health Centre during the same period. There was no case of death reported among the staff seen in either of the clinics during the period.

Table 1 shows the frequency distribution of all cases seen at the Staff clinic of the Teaching Hospital during the period. 844 of the cases were diagnosed as malaria giving a prevalence rate of 43% of all cases seen.

Table 2 shows the distribution of diseases seen at the Okelele health centre. 513 cases were due to malaria accounting for a prevalence rate of 36.7%. In both populations, the ratio of male to female was approximately 1:2.

During the period of study, a total of 51 Sick Leave Certificates were issued in the Staff Clinic for all cases, out of which 31 (60.8%) were due to malaria (Table 3). Table 4 shows the sickness absence (Sick Leave Certificates) due to malaria among the workers according to their occupations.

Table 1: Distribution of Diseases seen at the Staff Clinic of the University of Ilorin Teaching Hospital between August and October, 2001

Type of Diseases	August Frequency	September Frequency	October Frequency	Total	Percentage
Malaria	243	312	289	844	43.0
URTI	62	87	108	257	13.1
Gastroenteritis	6	20	21	47	2.4
Skin diseases	8	17	16	41	2.1
Eye/ENT diseases	10	6	25	41	2.1
UTI	3	2	3	8	0.4
Others	277	233	167	677	34.5
Referred cases	21	10	17	48	2.4
Total	630	687	646	1963	100

- URTI - Upper respiratory tract infection
 ENT - Ear, Nose and Throat
 UTI - Urinary tract infection.

Table 2: Distribution of Diseases seen at Okelele Health Clinic, Ilorin between August and October, 2001.

Diseases	August Frequency	September Frequency	October Frequency	Total	Percentage
Malaria	108	164	241	513	36.7
URTI	4	69	62	135	9.7
Gastroenteritis	0	11	6	17	1.2
Skin diseases	2	2	8	12	0.9
Eye Diseases	8	7	0	15	1.1
UTI	1	2	3	6	0.4
Others	116	245	289	650	46.5
Referred	9	19	21	49	3.5
Total	248	519	630	1397	100

URTI - Upper respiratory tract infection
 UTI - Urinary tract infection.

Table 3: Sick Leave Certificates issued at the Staff Clinic of the University of Ilorin Teaching Hospital during period of study.

Type of illnesses	Number of Sick Leave Certificates Issued				
	August	September	October	Total	Percentage
Malaria	8	10	13	31	60.8
URTI	3	3	4	10	19.7
Gastroenteritis	1	1	2	4	7.8
Eye/ENT					
Conditions	0	0	2	2	3.9
Others	1	1	2	4	7.8
Total	13	15	23	51	100

Table 4: Absenteeism rates among employees seen at the Staff Clinic of Ilorin teaching Hospital, Ilorin due to Malaria according to Occupational groups from August to October, 2001.

Occupational groups	Number of workers	Absence due to Malaria	Percentage due to Malaria
Doctors	8	4	50.0
Nurses	16	10	62.5
Pharmacists	6	3	50.0
Administrative staff	9	5	55.5
Skilled workers	4	3	75.0
Unskilled workers	8	6	75.0
Total	51	31	60.8

DISCUSSION

Malaria is endemic in Nigeria as in other tropical countries. Hence it would be expected that every individual in the country including workers should be at risk of contracting the disease at any time (6). However, the impact of malaria on productivity in Nigeria is yet to be determined independently. This study has shown that malaria takes almost the same morbidity rate among the workers studied (43%) compared with the control population (36.7%) hence is not, strictly speaking, an Occupational disease among this group of workers. However, it goes to show that the attending health workers in any Occupational Health Service should also be familiar with prevailing endemic diseases among workers they see.

It is however, on the other hand, clear from the study that Malaria is of Occupational health importance not only because it accounted for the highest disease prevalence but also because it was the commonest cause of sickness absence among the workers (60.8%). This is a significant finding because it means that Malaria could therefore be the greatest factor accounting for loss in working time in Nigeria. This same observation has been documented in a similar study in 1987 (3).

Hence if Malaria is not controlled adequately in Nigeria, productivity at work will continue to be low. This is also true of other West African countries (7).

The total number of days of absence from work for the 51 Sick Leave Certificates issued were calculated to be 179 days. Out of these, Malaria alone accounted for 87 days or 48.6% of days lost. This is an average of 2.8 days per spell of Malaria. Other diseases accounted for varying number of days. For example,

there was one case of Sickle Cell disease crisis, which accounted for 13 working days. While this might be considered a considerable length of time, it should be noted that this was just one case of that disease whereas Malaria cases were 31 in all. When this situation is looked into critically, therefore the contribution of Malaria, as a cause of morbidity among the health workers becomes very significant.

Furthermore, Malaria also accounted for 50 percent or more of sickness absence among each category of workers. Hence, 50 percent of the Doctors issued Sick Leave Certificates were ill due to malaria while it accounted for 62.5% among the Nurses. This finding is very noteworthy showing again that Malaria could also affect productivity at the highest level of health care delivery in the developing countries. By extension, it is also an indication that the disease could sap the highest level of manpower of tropical countries thereby responsible for low economic development.

Also in the study, it was found that the ratio of male to female studied in both populations was approximately 1:2. This observation means that more female workers make use of the Staff Clinic and also are responsible for more sickness absence for all diseases including Malaria than the male workers. This finding is similar to that of previous studies on sickness absence (3,8,9).

Among the workers issued Sick Leave Certificates, Nurses accounted for the highest number. Indeed, Nurses accounted for 10 out of the 31 cases of patients issued Sick Leave for malaria, that is about one-third of the patients. This finding supports a previous study by Pines et al., (8) in which Nurses recorded a similar high level of sickness absence.

Doctors and Nurses are very important in health care delivery and if they fall ill, the whole of the health system would be affected. Hence the

significance of the findings of this study and in particular the importance of Malaria as a cause of morbidity among these cadres of workers. The Government therefore needs to structure its malaria control programme to include especially the health workers. The need for an occupational health service for health workers has previously been emphasized (10).

Other diseases, which accounted for the remaining 57% of cases among the study group and 63.3% among the control group, included Upper Respiratory Tract Infections, Gastroenteritis, Skin diseases, Eye/ENT diseases, Urinary Tract infections. Among those cases listed as others in Table 1 were chronic diseases including Sickle Cell diseases, Systemic hypertension, Lumbago and peptic ulcers. Most of the cases seen were treated by the Doctors at the Staff Clinic while a few especially the chronic conditions (i.e. about 3.3% in August, 1.5% in September and 2.6% in October) were referred for specialized management.

However, all the cases of Malaria were treated at the Staff Clinic. This no doubt emphasizes the fact that Malaria, although a major cause of morbidity among the working class, is not normally a cause of significant mortality among them because being adults they have developed high levels of immunity against the disease (11).

CONCLUSION

Malaria, an endemic disease in Nigeria was the commonest cause of morbidity among health workers attending the Staff Clinic of the University of Ilorin Teaching Hospital. It was responsible for nearly two-thirds of sickness absence among the workers including Doctors and Nurses. The implication of this is that health personnel being absent from

work due to malaria could affect the health system of the country hence the health of the general public due to poor health care delivery. This could result in low productivity with resultant effect on the economic development of the country.

RECOMMENDATION

In view of the high prevalence of Malaria in Nigeria and its resultant deleterious effects on the working population and hence on the Economic activities of Nigeria, there is an urgent need to control Malaria in general. The following recommendation should assist to achieve this objective:

1. Clear-cut case definition for Malaria should be made available for health workers. This is to enable them to take care of themselves and also to cater for the general population.
2. The current WHO Malaria control programme tagged "Roll-back Malaria programme" should be incorporated into the Curricula of various schools including Primary, Secondary levels and those of Institutions such as Medical Schools and Schools of Nursing and other similar health Institutions.
3. Governments in the developing countries need to subsidize the cost of Insecticide Treated Bed nets (ITNS) to enable more people afford them.
4. There should be closer supervision of reporting of occurrence of Endemic Diseases in the country by the respective health authorities using standard forms (-DSN-Disease Surveillance and Notification forms) and sending them to the appropriate officers.
5. Education of the public including workers needs to be embarked upon on a continuous basis through Seminars and

Workshops in order for them to adopt preventive and health promotion strategies aimed at reducing the incidence of Malaria.

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PARASITIC DERMATOSES AS SEEN AT THE UNIVERSITY OF BENIN TEACHING HOSPITAL (UBTH), BENIN CITY IN NIGERIA

¹Airauhi, LU; ²Ohunu, AN ²Otabor, CU

Departments of ¹Microbiology, ²Medicine, School of Medicine, College of Medical sciences, University of Benin, Nigeria
Correspondence: DR LU AIRAUHI, Department of Medical Microbiology, School of Medicine, College of Medical Sciences, University of Benin, Nigeria.

The epidemiology and associated risk factors for parasitic infections causing dermatologic lesions were studied retrospectively over a five – year period (1993-1998) in Benin City Nigeria. The study population comprised one hundred and fifty six patients (84 males and 72 females) out of a total of 1665 patients who attended the dermatology clinic at the (UBTH) during the period of study. Dermatological manifestations of diagnosed parasitic infections were recorded and related to the occurrence of parasite species in microscopically studied specimens collected from patients. 9.4% of patients seen presented with various skin diseases of parasitic origin. The most prevalent parasitic disease seen was scabies 115 (73.7%); others were onchodermatitis 16(10.3%), myiasis 11(7.1%), wuchereriasis (elephantiasis) 9(5.8%), cutaneous larva migrans 3(1.9) and pediculosis pubis 2(1.3%). Infection was prevalent in all age groups. Overall prevalence revealed that patients aged 15 years and below had the highest infection rate of 60 (38.5%) while the lowest infection occurred among those who were aged 60 years and above. Dermatologic parasitoses presented as chronic persistent infections, which were sometimes severe especially in children. Infection rate was significantly higher among males (53.8%) than females (46.2%) ($P<0.05$), symptoms included generalized skin rashes, for most infections, leopard skin in onchodermatitis and marked discomfort and disfigurement in elephantiasis. The major risks associated with these parasitic infections include socioeconomic status, age and human behavioral factors.

Key words, Parasite, dermatologic lesions, Nigeria.

Running titles, Dermatologic manifestations of Parasitoses in BeninCity.

INTRODUCTION

Although the skin is a protective organ it frequently succumbs to the attack of microorganism, parasites and arthropods (1) it has been reported that the continuous exposure of the skin to the environment makes it a common portal of entry for most parasites (2). Such point of entry could be associated with local tissue reaction and dermatologic changes, besides cutaneous lesions, parasites can produce systemic diseases in humans (2).

Some parasite species documented in association with dermatologic lesions include *Onchocerca volvulus* in biopsied tissue studies in Benin city, Nigeria (3). Onchocerciasis has been studied extensively in Nigeria and an alternative diagnostic method for onchocerciasis other than skin snipping has been described (4), *Paragonimus westermani* was demonstrated in cutaneous lesions in Japans (5)

Other studies on parasitic dermatoses include cutaneous myiasis due to *Cordylobia anthropoga* in Ilorin (6); wound myiasis due to *Cochlomyia hominivorus* in Libya (7). Although these surveys represent significant advances in the research on parasitic dermatoses, no study on parasitic diseases manifesting dermatologic lesions has been conducted at the UBTH, Benin City, Nigeria.

We noted increasing cases of dermatologic manifestation of parasitic infections in our center and to support the development of a strategy for the control of parasitic diseases, we undertook this study, designed to document common parasitic dermatoses seen in Benin City their epidemiology and associated risk factors.

MATERIALS / PATIENTS AND METHODS
STUDY AREA

This study was conducted at the UBTH, which is a public population based hospital in Benin City, Nigeria with a well-defined catchment area.

The University of Benin Teaching Hospital (UBTH) Benin City is a 400-bedded tertiary hospital that caters for the needs of Edo and Delta State. It offers specialist services in various medical disciplines including Dermatology. It is located in the tropical rainforest belt with high humidity, high annual rainfall and has an average temperature of 32°C.

METHOD

Records of patients managed at the consultant outpatient Dermatology clinic over a six-year period from January 1993 and December 1998 for parasitic dermatoses were retrospectively analyzed. Information obtained from such records included personal biodata, clinical features and diagnoses. As a matter of routine, ectoparasites are treated overnight in 10% potassium hydroxide and examined microscopically as already described by Belding, 1964(8). Similarly biopsy tissues were processed in the Shandon Elliot automatic tissue processor, sectioned and stained in Haematoxylin and Eosin and subsequently examined microscopically.

Patients were classified into classes 1-5 on the basis of socioeconomic status of patients and of parents in cases of children from data available in the hospital records as earlier described (9):

1. Upper and middle class
2. Intermediate class
3. Skilled workers and Clerical workers class
4. Semi skilled workers class
5. Unskilled workers class

STATISTICAL ANALYSIS

For the purpose of analysis, the patients were subdivided into age groups as shown below

<u>Patients</u>	<u>Age in year</u>
Children	15 year and below
Young Adults	16 - 25
Adults	26 - 40
Middle Aged	41 - 59
Aged	60 years and above

The chi-square (χ^2) test was used in assessing the inter or intra group differences in the proportion of observations that were assessed. Probability of less than 0.05 was considered significant (10).

RESULTS

A total of 156 (9.4%) patients had parasitic infections from among the 1665 patients seen at the dermatology clinic during the period of study. Diagnosis of parasitic dermatoses was based on characteristic clinical features and microscopic identification of parasites recovered from the lesions. The following parasites were isolated from the samples submitted to the laboratory for investigation.

Sarcoptes scabiei species was identified based on the characteristic morphological features previously described by Soulsby, 1982(11). Larva of *Cordylobia anthropophaga* was recognized based on characteristic posterior spiracle (12) while *Pthirus pubis* was identified as described by Belding (8).

Helminth parasites were seen in histological skin sections of biopsies and were due to filarial worms namely: *Onchocerca volvulus*, these were seen as cross sections of the parasites revealing characteristic somatic muscles, cuticular annulations and other diagnostic features consistent with those already described(12). The other

filarial worm infection was due to *Wuchereria bancrofti* appearing characteristically revealing cuticular bosses and somatic muscles in tissue sections (12).

The majority of infections were due to *Sarcoptes scabiei* 73.7%. The pattern of infection for other parasite species encountered is as follows *Onchocerca volvulus* 10.3%, *Cordylobia anthropophaga* 7.1%,

Wuchereria bancrofti 5.8%, *Ancylostoma caninum* 1.9% and *Phthirus pubis* 1.3%. Thus *Sarcoptes scabiei*, *Onchocerca volvulus*, *Cordylobia anthropophaga* and *Wuchereria bancrofti* are important human parasite species manifesting dermatologic lesions in our center. The relative species prevalence of parasitic infection manifesting dermatologic lesions is presented in Table 1.

TABLE 1: RELATIVE PREVALENCE OF PARASITIC INFECTION MANIFESTING DERMATOLOGICAL LESIONS.

Causal Parasite Species	Disease class	Relative species prevalence		Overall relative species Prevalence
		Males	Females	No%
		No%	No%	No%
<i>Sarcoptes Scabiei</i>	Scabies	67(43.0 ^a)	48(30.8 ^a)	115(73.7)
<i>Onchocerca Volvulus</i>	Onchodermatitis	6(3.8)	10(6.4)	16(10.3)
<i>Cordylobia Anthropophaga</i>	Myiasis	5(3.2)	6(3.8)	11(7.1)
<i>Wuchereria Bancrofti</i>	Elephantiasis	6(3.8)	3(1.9)	9(5.8)
<i>Ancylostoma Caninum</i>	Cutaneous larva migrans	0(0.0)	3(1.9)	3(1.9)
<i>Phthirus pubis</i>	Pediculosis pubis	0(0.0)	2(1.3)	2(1.3)
Total		98(53.8 ^a)	72(46.2 ^a)	156(100)

^aValues differ significantly from each other p<0.05

The prevalence of *Sarcoptes scabiei* increased with decreasing age in both sexes with an overall prevalence of scabies of 43.0% in males and 30.8% in females. The incidences of infection as they occurred in the various age groups were expressed in absolute numbers and percentages. Of the 73.7% prevalence of scabies, 19.2% and 12.8% were from males and females respectively among patients aged 0 – 15 years old.

Filariases were due to *Onchocerca volvulus* and *Wuchereria bancrofti*. Of the 16(10.3%) cases of onchocerciasis 6(3.8%) and 10(6.4%) were recorded among male and female subjects respectively. The pattern of onchocerciasis reveals that infection was recorded among males aged between 41 – 49 years while for females infection occurred among subjects aged between 26 and over 60 years of age. The prevalence pattern of infection for wuchereriosis is similar to that for onchocerciasis in males while for

females a prevalence infection rate of 3(1.9%) was recorded among subjects aged 60 years and above.

The overall prevalence of cutaneous myiasis due to *Cordylobia anthropophaga* was 11(7.1%), of this 5(3.2%) were seen in males aged 0-15 years of age, in females a prevalence

4 (2.6%), 1(0.6%) among those aged 26-40 years and 41-59 years respectively. The number and parasite species seen in-patients according to age and sex are presented in Table 2.

Patients from lower social class were significantly more infected than those from higher class ($p < 0.05$).

TABLE 2: No. and parasite prevalence (%) in patients seen at the dermatology clinic by sex and age in Benin City, Nigeria, 1993-1998.

Ages (years)	No of patients seen							
	Cases Seen	No with parasitic Disease	<i>S. scabiei</i>	<i>O. volutulis</i>	<i>C. anthropophaga</i>	<i>W. bancrofti</i>	<i>A. caninum</i>	<i>P. pubis</i>
Males								
0-15	139	35(22.4)	30(19.2)	-(0.0)	5(3.2)	-(0.0)	-(0.0)	-(0.0)
16-25	151	16(10.3)	16(10.3)	-(0.0)	-(0.0)	-(0.0)	-(0.0)	-(0.0)
26-40	146	10(6.4)	10(6.4)	-(0.0)	-(0.0)	-(0.0)	-(0.0)	-(0.0)
41-59	140	19(12.2)	7(4.5)	6(3.8)	-(0.0)	6(3.8)	-(0.0)	-(0.0)
>60	156	4(2.6)	4(2.6)	-(0.0)	-(0.0)	-(0.0)	-(0.0)	-(0.0)
Total	734	84(53.8)	67(43.0)	6(3.8)	5(3.2)	6(3.8)	-(0.0)	-(0.0)
Females								
0-15	186	25(16.0)	20(12.8)	-(0.0)	4(2.6)	-(0.0)	1(0.6)	-(0.0)
16-25	178	13(8.3)	11(7.1)	-(0.0)	-(0.0)	-(0.0)	-(0.0)	2(1.3)
26-40	194	12(7.7)	7(4.5)	4(2.6)	1(0.6)	-(0.0)	-(0.0)	-(0.0)
41-59	155	11(7.1)	7(4.5)	3(1.9)	1(0.6)	-(0.0)	-(0.0)	-(0.0)
>60	218	11(7.1)	3(1.9)	3(1.9)	-(0.0)	3(1.9)	2(1.9)	-(0.0)
Total	931	72(46.1)	48(30.8)	10(6.4)	6(3.8)	3(1.9)	3(1.9)	2(1.3)
Overall Total	1665	156	115(73.7)	16(10.3)	11(7.1)	9(5.8)	3(1.9)	2(1.3)

* Values differ significantly from each other $p < 0.05$

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The clinical features of the dermatological lesions due to parasitic infections reveals that of the 115 patients with scabietic infection, 4(2.7%) males and 13(8.3%) females presented with generalized papular lesions; papular squamous scaly lesions were seen in 12 (7.7%) in both males and females while vesicular lesions were seen in 21(13.5%) males and 18(11.5%) in females, pustular lesions were seen in 19(12.2%) in males and 20(12.8%) in females. of 6(3.8%) was recorded, occurring among subjects aged 0-15 years with

DISCUSSION

The pattern of parasite species manifesting dermatologic lesion in Benin City, Edo State is demonstrated in this study. Scabies was the most prevalent dermatologic parasitic infection with an overall prevalence of 73%, with a significantly higher prevalence among males compared to females ($P < 0.05$). We are not able to explain this pattern of infection, we therefore suggest that this finding should be interpreted with caution. The highest infection was recorded among patients aged 0-15 years. Thus a possible interpretation of

this result is that children are most infected which corroborates other reported findings that children are more at risk of acquiring scabietic infection (13,14). This however varies from observation in Zaria, Nigeria from where age dependent pattern of infection were reported not to exist (15). It would seem that scabies being a contagious disease is most likely spread in children when at play, sleep and other interactions at home by direct contact. Our finding of increasing scabietic infection with decreasing age can therefore not be taken as an accepted general pattern of scabietic infection. This association is most likely a chance occurrence, especially when overcrowding is well documented as an important predisposing factor for scabietic infection (14,15). The variation in prevalence rates from various studies may be due to variations in study populations. On the other hand it could also be that the high prevalence recorded in childhood population may be an indication of slow acquisition of resistance in children. An earlier explanation for decreasing prevalence rate of scabies with advancing age may be the presence of acquired immunity to previous clinical and sub-clinical infection(15)

Several prospective studies have shown that onchodermatitis and elephantiasis are manifestations of long standing filariasis (16,17,18) although these findings cannot be said to be conclusive. We however observed that filariasis due to *Onchocerca volvulus* and *Wuchereria bancrofti* were recorded among adults, middle aged and aged population. Our reported pattern thus provides support for these earlier findings that suggests chronicity of filarises. This concept is a strong indication that onchodermatitis and elephantiasis are manifestation of long standing filariasis. The overall prevalence of

onchodermatitis and elephantiasis was 10.3% and 5.8% respectively (Table2).

We reported 7.1% cases of myiasis due to *Cordylobia anthropophaga* larvae with 5(3.2%) and 4(2.6%) from males and females respectively among children. In the childhood population children less than 5 years old were most infected which is consistent with earlier reports (5,17). As suggested earlier the high prevalence of myiasis in children less than five years may be partly due to neglect and also partly due to behavioural pattern among children (19). Children have been reported to be left unchanged out of their soiled clothes for long hours while playing in the yards, often soiled in their urine and excrements. Most children in rural settings do not have access to proper toilet facilities and those in semi urban settings are not toilet trained. These therefore expose them to constant risks for infections, thus neglect of children and poor toilet habits have emerged as factors to consider in the epidemiology of myiasis. The dirty environment thus created by poor standard of living and neglect serve as attractant for female *Cordylobia anthropophaga* flies, which lay eggs that hatch into the offending tissue invading larvae. Considering the consistency of demonstrated association in age and relationship to the prevalence of myiasis, it is likely that behavioral habits and neglect of children contribute to the pattern of infection reported in children in this study.

1 (0.6%) case of cutaneous larva migrans was recorded in a female child aged less than 1 year and 2 (1.3%) case in 2 female aged over 60 years. Cutaneous larva migrans due to invasion by larvae of *Ancylostoma caninum* is a zoonosis, dogs stray and defecate around yards where children play partly naked and adults walk barefooted thus creating an enabling environment for the offending larvae to invade by the percutaneous route.

2 (1.3%) cases of Pediculus pubis were recorded in female aged between 16-25

years. One of these patients had been raped although we are not able to confirm if this infection was present before the rape, it is however worthy of note that one of the subjects had been sexually assaulted.

Both males and females were characterized by noteworthy multiple manifestation of dermatologic lesions in the same parasitoses. In this study we confirm a significantly higher occurrence of multiple lesions ($p < 0.05$) and wider spread of lesions ($p < 0.05$) in scabietic and onchocercal lesions compared to wuchereriosis, myiasis, cutaneous larva migrans and pediculosis as previously documented (5,6,15,16). There were no sex dependent significant group differences in clinical manifestations of lesions. Our report of higher occurrence of multiple and wide spread lesions of scabies and onchodermatitis is consistent with earlier recorded findings (20). The non sex dependent significant differences in clinical manifestation may possibly be attributed to the fact there is no sex predilection in clinical manifestations of infections. The face was characteristically spared in scabies this provides support for an earlier report that scabietic lesions do not occur in skins above the neckline because the etiologic parasite avoids areas with cold temperatures. (19).

A significantly higher scabietic prevalence of infection was observed among patients from the lower social class ($p < 0.05$) which provides support for an earlier reported finding that factors like overcrowding and poor hygiene may promote parasitic dermatoses particularly scabies (14,15). Social class is therefore a factor of importance in the transmission and spread of infection.

We recommend that the best way to ensure cure rate in parasitic dermatoses is early recognition of lesions. Parasitologic confirmation of the offending parasite species may be

needed before treatment is commenced. This will help to reduce the spread of lesions and associated morbidities. The effect of elephantiasis as observed was that infected people developed functional incapacities that increased their dependence on family members with its associated economic impact, as sufferers became unproductive.

This work highlights the significance of studies of dermatologic manifestations of parasitic diseases. Information provided here will serve to facilitate case detection and improved quality care through enhanced clinical skills.

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PROFILE OF POTENTIALLY PATHOGENIC INTESTINAL PARASITES AND BACTERIAL AGENTS IN SOLID WASTES IN IBADAN MUNICIPALITY.

BY: *ADEYEBA, O.A & AKINBO, J.A, *Department of Medical Microbiology & Parasitology College of Health Sciences, Ladoko Akintola University of Technology, PMB 4400, Osogbo, Nigeria & Department of Medical Microbiology and Parasitology, University of Ibadan, Ibadan, Nigeria

In order to determine the profile of potentially pathogenic enteric parasites and bacterial agents in municipal refuse dumps in Ibadan, 5 major market places in the city were randomly selected by balloting method. Refuse sludge was examined parasitologically and bacteriological using the method described. The data analysis was done and test of significance carried out by using the chi square test where applicable.

Cases of multiple parasites and bacterial agents were commonly encountered in the sludge refuse samples.

The commonly found parasitic agents were of both human and veterinary importance. These included *Ascaris lumbricoides* (9.3 egg), *Entamoeba histolytica* (8.07 cyst per gram); Hookworm *strongyle* (6.27 egg) and *Ascaris suum* (1.07 egg). Others are *Ascaris vitolorum* (1.09 egg) *Stongyloides papillosu* (0.52 larva/egg) *Schistosoma suis* (0.31 egg) and *Dicrocoelium dendriticum* whilst the most commonly found bacterial agents were *Klebsiella* species, *Escherichia coli*, *Proteus* specie, streptococci and other gram-positive organisms. Climatic conditions affected the distribution of both parasites and bacterial agents in the sludge ($P < 0.001$). more intestinal parasites (53.4%) and bacterial agents (27.2%) were encountered at mean air temperature $26.1 \pm 0.6^{\circ}\text{C}$, mean relative humidity of $72 \pm 3.5\%$. The degree of contamination by market location varies significantly ($P < 0.001$) A high degree of contamination of solid waste dumpsites with bacterial and parasitic agents was observed in the present study. As a result of the public importance of the organisms isolated it is opined that well planned waste management and health education programs will go a long way to reduce the potential epidemic risks posed by such sites in Ibadan, Nigeria. it is believed that economic advantage could be taken of the mountainous solid waste dump by establishing fertilizer-processing plant to produce fertilizer for farmers and provide job opportunity for youths in the area.

KEY WORDS: Pathogens, Contamination, Intestinal Parasites, Bacteria, Refuse Dump, Markets, Ibadan, Health Education, BeninCity.

INTRODUCTION

Refuse, soil, animal waste and sewage sludge are common sources of manure, used to fertilize agriculture fields (1,2,3). Studies have revealed the incidence and distribution of many pathogenic intestinal parasites and bacterial agents from refuse which infect both man and animals (4,5,6). The most commonly found bacterial agents include gram-negative enteric bacteria like *Pseudomonas* species, *Salmonella* species, *Klebsiella* species, *Escherichia coli*, *Aeromonas* species and some gram-positive organisms (4,5,6,7,8).

Similarly intestinal parasites are life in many communities and are of such a major international health concern (9). It has been shown that refuse dumps are significant sources of transmission for intestinal parasitic infection in Kampala,

Uganda and Jos, Nigeria (10,11). Whereas many workers have isolated veterinary and medically important parasitic agents from refuse dumps and abattoir in some parts of the world (11,12,13), there is dearth of information on the status of refuse dumps in Southwestern Nigeria. Hence this study is designed to determine the profile of potentially pathogenic enteric bacterial and parasitic agents in refuse dumps in major markets in Ibadan metropolis.

MATERIALS AND METHODS

Study Area: This study was carried out between July 1999 and January 2000 in Ibadan, capital of Oyo State, Nigeria; the most populous black city south of the Sahara Desert with about 3.5 million inhabitants (1991 national census). Being a metropolitan city, there is influx

of people of other nationalities beside the local indigenes for many reasons including socio-economic and political. This deluge of people into the city has resulted in the proliferation of markets in the city to the extent that numerous refuse dumps are common features because of the inability of the responsible authority to cope with the disposal. It is common to see refuse dumps disturbing both vehicular and pedestrian movements in the city. Ibadan experiences both wet and dry seasons. April and October span the rainy season with average rainfall of about 550mm and 1000mm while the dry season cover the period of November and March (Federal office statistics, Lagos, 1998).

Sample Selection: For the purpose of this study 5 major market places were randomly selected (by balloting method) from the list of markets in the metropolis.

Sample Collection: A total of 1610-refuse sludge samples was collected in all the refuse dumps in the study areas. Each of the 5 refuse locations was visited 14 times and the collections were done twice in the day between 7.00hr-8.00hr (mean relative humidity of 82± 3.0%; mean air temperature of 26.1 ±0.6°C) and 15.00hr - 17.00hr (mean relative humidity of 72 ± 3.5%; mean air temperature of 28.4±0.5°C) respectively. On each of the first and second visit, 23 samples were taken to make up 322 samples taken from each refuse locations at the end of 7 months period of the study.

About 100g each sample of sludge taken from a depth of 10cms from each refuse site was collected with the aid of sterile wooden spatula into sterile 20 cc screw - capped plastic bottle. This was done in order to ensure uniformity in sampling. Samples from each site were later

pooled together to produce a single sample for analysis dumpsite.

Sample Analysis:

a. **Parasitology:** This was carried out by using the technique described by Kegei (14). 100g refuse sludge as weighed was passed through a coarse sieve of 4mm² pore size to remove stones, grass and other undesirables. The preparation was later transferred quantitatively to 50ml volumetric flask. To each volume of refuse, 2 volumes of 30% sodium hypochlorite fluid were added as disinfectant, vigorously stirred and allowed to stand for 30 minutes. This mixture was further diluted to the 50ml mark and mixed again. Coarse particle was stained out by passing through a coarse mesh clothe into a centrifuge tube and centrifuged at 3000 rpm for 2 minutes. The supernatant was discarded and the deposit re-suspended in magnesium sulphate floatation fluid of specific gravity 1.3, and then centrifuged again at 3000 rpm for another 2 minutes.

The floatation fluid in the centrifuge tube was then filled up to form a positive meniscus and a coverslip was superimposed on it and left to stand for 5 minutes. The coverslip was then lifted with a swift action and placed on a glass and examined microscopically for the presence of cyst and eggs of parasite as described by Lelano, *et al* (13).

b. **Bacteriology:** 100g refuse sludge was passed through a coarse sieve of 4mm² pore size to remove stones etc. this was homogenized in 50ml volumetric flask with sterile normal saline. Sample was seconded on agar, Maconkey agar and Decxycholate agar plates and Selenite Froth for the primary isolation of organism as described by Cruickshank, *et al* (15). All cultures were incubated at 37°C overnight in the presence of free oxygen. All isolates were characterized and identified by using the criteria of Cowan and Steel (16). Oxford *Staphylococcus* (NCTC 6571)

and *Escherichia coli* (CTC 10418) were used as control.

Data Analysis

The data analysis was done by using the computer and the chi-square test level of significance where applicable.

RESULT

Parasites encountered in refuse dumps in Ibadan. Table 1 shows the profile of the intestinal parasites isolated from refuse waste dumps in Ibadan metropolis with corresponding probable hosts. These parasites include protozoan cysts. Out of the oocyst and Helminthes eggs of man, dog, sheep pig, cattle and goat of the 18 different species recovered, protozoa cyst were 5, helminthes eggs 12 and flagellate 1. *Ascaris lumbricoides* (9.37 epg) is the most frequently encountered parasite, followed by *Entamoeba histolytica* (8.07 cysts) Hookworm/Strongyle (6.27 epg while *Schistosoma suis* (0.31 epg) was the least encountered.

Prevalence of intestinal parasite in Ibadan municipal refuse dump by market locations is shown in table 2. Result shows multiple parasitic agents in each refuse dump sample. The level of contamination with parasitic agents by refuse dump differ significantly ($X^2 = 1391.52$, $df = 4$, $P < 0.001$) of the total 4727 cases of polyparasitism by market locations, Sango, 1263 (26.7%) has the highest prevalence rate of multiple parasitic contamination. This was followed by Oritamerin market dump site 1245 (26.3%), Bodija, 1182 (25.0%), Oje, 599 (12.7%) and Dugbe, 438 (9.3%). Though Sango refuse dump appeared to be the most contaminated of all, this variation, compared to Oritamerin and Bodija refuse location was statistically significant ($P < 0.001$)

Table 3 shows the prevalence of intestinal parasites encountered in

Ibadan refuse dumps at mean air temperature 26.1 ± 0.6 and mean relative humidity of $82 \pm 3.0\%$. Results show the profile of intestinal parasite encountered. Data revealed the degree of contamination's in each of the refuse locations of the total (2523) number of polyparasitism at this climatic condition, *A.lumbricoides* 20.2% had the most prevalent rate followed by *E. histolytica* 16.5%, strongyle 15.7% and *S.suis* 0.6% there is a statistical difference in the degree of contamination by refuse location ($X^2 = 1781.04$, $df = 4$, $P < 0.001$).

Table 4 shows the prevalence rate of intestinal parasites encountered at mean air temperature $28.4 \pm 0.5^\circ\text{C}$ and mean relative humidity $72 \pm 3.5\%$ of the 2204 positive cases of polyparasitism, *A.lumbricoides* (19.3%) had the highest prevalence rate, followed by *E.histolytica* (17.7% Hookworm (strongyle) 10.4% and *Dicrocoelium dendriticum* (0.4%) successively. The variations by refuse dump differ significantly ($X^2 = 98.61$, $df = 4$, $P < 0.001$). However, the difference Bodija (27.5%) and Oritamerin (28.7%) was not significant ($P > 0.001$).

The prevalence of the potentially pathogenic bacterial agents isolated from municipal dumps in Ibadan by market locations is shown in Table 5. The result shows a varying degree of multiple bacterial agents by each refuse location; oritamerin, 945 (25.1%) was the most frequently contaminated followed by Bodija, 882 (23.4%); Dugbe 755 (20.1%) and Sango 579 (15.4%). These variation shows a significant statistical difference ($X^2 = 2501.99$, $df = 4$, $P < 0.001$). However the difference between Sango 579 (15.4%) and Oje 601 (15.9%) was not significant ($P > 0.001$).

Table 6 shows the prevalence of potentially pathogenic enteric bacterial agents asserted at mean air temperature 26.1 ± 0.6 and mean relative humidity $82 \pm 3.0\%$ of the 2150 multiple bacterial agents isolated at this climatic

condition, *Klebsiella* specie 573 (26.7%) was the most prevalent followed by *E coli* 473 (22.0%) while the least encountered was *Streptococcus* species 12 (0.6%). There was significant difference in the degree of contamination by marker location ($X^2 = 1173$, $df=4$, $P<0.001$).

The prevalence of potentially pathogenic bacterial agents isolated at mean air temperature 28.4 ± 0.5 and mean humidity of $72 \pm 3.5\%$ of the 7612 cases of multiple bacterial contamination at this climatic condition, *E coli* 238 (14.8%) was the highest prevalent followed by *Klebsiella* specie 218 (13.8%) and *Streptococcus* specie 15 (0.9%). The variations in the contamination rate by refuse dump were also significant ($X^2 = 834.23$ $df =4$, $P<0.001$).

Table 1: PARASITIC OVA, CYSTS AND LARVAE FOUND IN REFUSE DUMPS IN IBADAN.

Parasite	Possible host	Total numbers of cyst/egg/100g
<i>Entamoeba histolytica</i>	man	807
<i>Entamoeba coli</i>	man	204
<i>Blantidium coli</i>	man	34
<i>Giardia lamblia</i>	man, dog, cat	190
<i>Trichomonas hominis</i>	man	506
Cocida(oocyst)	sheep	120
<i>Ascaris himbricoides</i>	man, pig	937
<i>Ascaris suum</i>	man, pig cattle	107
<i>Ascaris vitulorum</i>	cattle	109
Hookworm	man	627
<i>Trichuris trichiura</i>	man	160
<i>Trichuris ovis</i>	sheep, goat	333
<i>Strongyloides stercoralis</i>	man	66
<i>Trichostrongylus</i>	sheep	52
<i>Fasciola hepatica</i>	man, sheep, goat	202
<i>Teania specie</i>	man,	81
<i>Dicrocoehum dendriticum</i>	sheep, goat	48
<i>Schistosoma suis</i>	pig	31

Table 2: PREVALENCE OF INTESTINAL PARASITES IN MUNICIPAL REFUSE DUMP IN IBADAN BY MARKETS LOCATIONS. PERCENTAGE OF POSITIVE CASES.

Oje	Dugbe	Orita-merin	Sungo	Bodiji	Refuse location No. of samples
32.2	32.2	32.2	32.2	32.2	
24.2	24.2	10.1	26.9	10.7	<i>Entamoeba Histolytica</i> (%)
1.0	4.3	5.8	7.1	1.4	<i>Entamoeba coli</i> (%)
0	0	0	1.2	1.6	<i>Balanidium coli</i> (%)
4.3	8.6	4.3	3.9	2.0	<i>Giardia lamblia</i> (%)
8.3	13.9	14.41	11.2	6.2	<i>Trichostrongylus hominis</i> (%)
0	0	0.5	1.3	9.1	<i>Coccidia Oocyst</i> (%)
17.0	30.3	18.4	17.0	21.8	<i>Ascaris lumbricoides</i> (%)
0	0	3.3	0.7	4.8	<i>Ascaris Suum</i> (%)
6.8	0	4.3	0	1.2	<i>Ascans vitulo</i> (%)
14.6	17.1	14.3	16.8	6.6	<i>Hookworm</i> (%)
12.1	0	3.1	1.9	2.1	<i>Trichuris trichiurum</i> (%)
0	0	0.6	2.2	6.2	<i>Trichuris ovis</i> (%)
8.0	2.7	7.1	9.4	5.6	<i>Strongyloides stercoralis</i> (%)
0	0	1.8	0	3.7	<i>Trichostrongyl</i> (%)
0	0	0	0	4.4	<i>Strongyloides papillosum</i> (%)
0	1.5	8.5	0	7.5	<i>Fasciola hepatica</i> (%)
3.3	4.7	3.2	0	0	<i>Taenia spenic</i> (%)
0	0	0.9	0	3.1	<i>Dirocoelium ctenodactylum</i> (%)
0	0	0	0.7	1.9	<i>Schistosomusis</i> (%)
599	438	1245	1263	1182	Total with polyparasitism per dump site
12.7	9.3	26.3	26.7	25.0	Degree of contamination #

Table 3: PREVALENCE OF INTESTINAL PARASITE IN IBADAN MUNICIPAL REFUSE DUMPS AT MEAN AIR TEMPERATURE OF $26.1 \pm 0.6^\circ\text{C}$ AND MEAN RELATIVE HUMIDITY OF $82 \pm 3.0\%$.

PERCENTAGE OF POSITIVE CASES.

Oje	Dugbe	Orin-merin	Sango	Bodij	Refuse location
161	161	161	161	161	No. of samples
12.0	12.5	4.2	38.6	7.4	<i>Entamoeba histolytica</i> (%)
0.3	5.9	4.2	3.0	1.6	<i>Entamoeba coli</i> (%)
0	0	0	2.2	1.4	<i>Balan tidumkali</i> (%)
7.1	12.5	5.2	5.0	3.3	<i>Giardia lamblia</i> (%)
3.3	12.8	12.1	12.8	4.7	<i>Trichomonas hominis</i> (%)
0	0.7	0	0	8.7	<i>Coccidia Oojet</i> (%)
24.0	26.4	21.4	13.1	22.5	<i>Ascaris lumbricoides</i> (%)
0	0	0	0.4	3.6	<i>Ascaris Suam</i> (%)
5.5	0	5.2	0	0.7	<i>Ascans vitulo</i> (%)
19.1	18.3	13.9	20.8	8.2	Hookworm (%)
19.1	0	5.1	0.9	1.0	<i>Trichuris trichiura</i> (%)
0	0	0	1.4	7.6	<i>Trichuris ovis</i> (%)
6.8	4.4	11.3	1.1	2.4	<i>Strongyloides stercoralis</i> (%)
0	0	1.3	0	5.0	<i>Trichostrongyl</i> (%)
0	0	0	0	6.4	<i>Strongyloides papillosu</i> (%)
0	1.5	9.6	0	8.2	<i>Fasciola hepaticu</i> (%)
1.9	5.1	4.4	0	0	<i>Toxaria spece</i> (%)
0	0	2.0	0	4.7	<i>Dicrocoelium dendriticum</i> (%)
0	0	0	0.6	2.3	<i>Schistosomatus</i> (%)
366	273	62	696	576	Total Parasitism in refuse sample

Table 4: PREVALENCE OF INTESTINAL PARASITES IN IBADAN MUNICIPAL REFUSE DUMPS AT MEAN AIR TEMPERATURE OF $28.4 \pm 0.5^{\circ}\text{C}$ AND MEAN RELATIVE HUMIDITY OF $72 \pm 3.5\%$.

PERCENTAGE POSITIVITY

Oje	Dugbe	Orin-mann	Sango	Bodije	Refuse location
161	161	161	161	161	No. of samples
43.3	21.8	15.8	12.5	13.5	<i>Entamoeba histolytica</i> (%)
2.1	1.8	7.3	12.2	1.3	<i>Entamoeba coli</i> (%)
0	0	0	0	1.8	<i>Balantidium coli</i> (%)
	2.4	3.3	2.5	0.8	<i>Giardia lamblia</i> (%)
16.3	15.8	16.6	9.3	7.8	<i>Trichomonas hominis</i> (%)
	0	0	2.8	9.4	<i>Coccidia (Oocyst)</i> (%)
6.0	37.0	15.5	21.9	21.1	<i>Ascaris fulvicornis</i> (%)
9.0	0	3.5	0	5.9	<i>Ascaris suum</i> (%)
6.8	0	4.3	0	1.2	<i>Ascaris vitillo</i> (%)
7.7	15.2	14.7	10.9	5.3	<i>Hookworm</i> (%)
	0	1.1	3.2	3.1	<i>Trichuris trichiura</i> (%)
0	0	1.1	3.2	4.1	<i>Trichuris ovis</i> (%)
9.9	0	3.0	11.9	8.6	<i>Strongyloides stercoraris</i> (%)
0	0	2.2	0	2.5	<i>Trichostrongylus</i> (%)
0	0	0	0	2.5	<i>Strongyloides papillaria</i> (%)
0	1.8	7.7	0	6.5	<i>Fasciola hepatica</i> (%)
5.6	4.2	2.1	0	0	<i>Taenia sp.</i> (%)
0	0	0	0	1.5	<i>Dicrocoelium dendriticum</i> (%)
0	0	0	0.9	1.5	<i>Schistosoma</i> (%)
233	165	633	567	606	Total Parasites in refuse sample
10.6%	7.5%	38.7%	25.7%	27.5%	Degree of contamination

- * Total depict polyparasitism in refuse sample
- # Percentage contamination per refuse dump
- () Percentage contamination.

Table 5: INCIDENCE OF POTENTIALLY PATHOGENIC BACTERIA IN MUNICIPAL REFUSE DUMPS IN IBADAN BY MARKET LOCATIONS

PERCENT POSITIVITY

Refuse locations	No. of sample examined	<i>Staphylococcus aureus</i> (%)	<i>Staphylococcus albus</i> (%)	<i>Staphylococcus</i> Species (%)	<i>Yeast Cells</i> (%)	<i>Citrus Pasteure bacilli</i> (%)	<i>Klebsiella coli</i> (%)	<i>Klebsiella</i> specie (%)	<i>Proteus</i> specie (%)	<i>Pseudomonas</i> specie (%)	<i>Salmonella</i> specie (%)	Total *	Degree of contamination #
Bodiya	322	(5.6)	(10.3)	(2.6)	(5.2)	(11.5)	(17.4)	(20.6)	(6.0)	(10.9)	(4.6)	882	23.4%
Sango	322	(3.2)	(2.7)	(0.6)	(9.7)	(15.0)	(15.8)	(23.4)	(7.1)	(10.9)	(5.1)	579	15.4%
Orta merrn	322	(7.1)	(3.8)	0	(3.8)	(9.3)	(10.4)	(25.2)	(10.4)	(19.7)	(1.2)	945	25.1%
Dugbe	322	(8.2)	(6.1)	0	(3.8)	(14.8)	(20.7)	(16.1)	(11.1)	(9.9)	(9.0)	755	20.1%
Oje	322	(6.4)	(4.2)	0	(1.9)	(19.4)	(10.9)	(18.6)	(11.8)	(15.8)	(0.6)	601	15.9%

* Total depict polyparasitism in refuse sample
 # Percentage contamination per refuse dump

Table 6: PREVALENCE OF POTENTIALLY PATHOGENIC BACTERIA IN REFUSE DUMPS IN IBADAN AT MEAN AIR TEMPERATURE OF 26.1 ± 0.6°C AND MEAN RELATIVE HUMIDITY OF 82 ± 3.0%.

PERCENT POSITIVITY

Refuse locations	No. of sample examined	<i>Staphylococcus aureus</i> (%)	<i>Staphylococcus albus</i> (%)	<i>Staphylococcus</i> Species (%)	Yeast Cells (%)	Gram Positive bacilli (%)	<i>Escherichia coli</i> (%)	<i>Klebsiella</i> species (%)	<i>Proteus</i> species (%)	<i>Pseudomonas</i> species (%)	<i>Salmonella</i> species (%)	Total *	Degree of contamination
Bodija	161	(4.1)	(2.3)	(2.3)	(6.4)	(10.9)	(22.2)	(17.3)	(2.8)	(12.4)	(4.9)	532	24.2%
Sango	161	(5.1)	(1.2)	0	(10.6)	(16.0)	(15.4)	(27.8)	(3.3)	(9.7)	(4.5)	331	15.1%
Orimerrin	161	(4.8)	(1.3)	0	(2.6)	(7.9)	(1.7)	(31.8)	(10.3)	(18.6)	(1.1)	544	25.3%
Dugbe	161	(5.4)	(1.0)	0	(6.2)	(16.7)	(25.1)	(23.8)	(17.2)	(3.8)	(0.8)	390	18.1%
Oje	161	(5.9)	(2.5)	0	(0.8)	(21.5)	(24.9)	(19.8)	(15.3)	(8.5)	(0.6)	353	16.4%

* Total depict polyparasitism in refuse sample

Percentage contamination per refuse dump

Table 7: PREVALENCE OF POTENTIALLY PATHOGENIC BACTERIA IN REFUSE DUMPS IN IBADAN AT MEAN AIR TEMPERATURE OF $28.4 \pm 0.5^{\circ}\text{C}$ AND MEAN RELATIVE HUMIDITY OF $72 \pm 3.5\%$.

PERCENT POSITIVITY

Refuse locations	No. of sample examined	<i>Staphylococcus aureus</i> (%)	<i>Staphylococcus albus</i> (%)	<i>Staphylococcus Specae</i> (%)	Yeast Cells (%)	Gram Positive bacilli (%)	<i>Escherichia coli</i> (%)	<i>Klebsiella specae</i> (%)	<i>Proteus specae</i> (%)	<i>Pseudomonas specae</i> (%)	<i>Salmonella specae</i> (%)	Total *	Degree of contamination #
Bodija	161	(7.7)	(17.1)	(3.7)	(3.4)	(12.3)	(10)	(10.6)	(10.9)	(10.6)	(1.3)	350	21.7%
Scungo	161	(0.8)	(4.8)	(1.6)	0	(13.7)	(6.5)	(17.7)	(12.1)	(6.6)	(6.1)	248	15.4%
Orta- merin	161	(11.2)	(11.5)	0	(5.5)	(11.2)	(16.2)	(16.4)	(10.5)	(11.4)	(1.2)	401	24.8%
Dugbe	161	(11.2)	(11.5)	0	(1.4)	(12.7)	(16.2)	(7.9)	(4.7)	(16.4)	(17.8)	365	22.6%
Oje	161	(7.3)	(6.5)	0	(3.5)	(16.5)	(15.3)	(16.9)	(6.9)	(17.8)	0	365	22.6%

* Total depict polyparasitism in refuse sample
 # Percentage contamination per refuse dump

DISCUSSION

This study has shown that there is a high degree of refuse contamination with pathogenic human animal intestinal parasites and bacterial agents in Ibadan municipality. The commonly found intestinal parasites include *A lumbricoides*, *E histolytica*, hookworm (strongyle). While the least encountered was *Schistosoma suis*. The cysts, oocyst and helminthes eggs recovered from the refuse dump sample were essentially those that were shed in the faeces of human and animal which became dispersed indiscriminately to refuse dumps. Other potential sources include litters from poultry farms, piggeries, sheep, goat market in the study area and waste from abattoir houses. These sources of cysts and eggs in refuse dumps are similar to those previously reported elsewhere in Nigeria (1,17) and in other part of the world (18,19). The report of isolation of intestinal parasites of veterinary importance such as *A suum.*, *a. vitulorum* and *Sreonyloides papillosu* in Ibadan refuse dumps accords well with the report of Burger (12) that intestinal parasites of veterinary importance are capable of being transmitted to the public through abattoir waste which were indiscriminately deposited in the refuse dump.

The isolation of these parasitic agents from municipal refuse is highly as the parasites are capable of causing outbreak of water or food borne amoebiasis, giardiasis or balantidiasis through the contaminative route (11). And this is in consonance with reports of outbreak of giardiasis from cyst and oocyst in municipal sludge (18,19). These reports corroborate the fact that other parasitic agents isolated in this study are potential courses of infection to the population in Ibadan.

This study has shown that there were cases of multiple parasitic contamination in each refuse dump sample. The prevalence of the parasites by market locations has shown a statistically significant difference ($X^2=1391.52, df=4, P<0.001$). This meant that risk of contacting disease in these areas is relatively the same. The level of pollution in these market places is found to be higher than the rest as residential buildings in the area have no toilets nor waste disposal facilities often use the market as dumping ground for their excrement and other wastes.

It has been shown in this study that climatic condition has significant impact on the occurrence rate of parasitic agents in refuse dump in Ibadan. More intestinal parasites (53.4%) were isolated at mean air temperature $26.1 \pm 0.6^\circ\text{C}$ mean relative humidity of $8.2 \pm 3.0\%$ than 46.6% isolated at mean air temperature 28.4 ± 0.5 and mean relative humidity $72 \pm 3.5\%$. This report accords well with other reports (20,21,22) that the survival of intestinal parasites is dependent on favourable degree of temperature, moisture, humidity, desiccation, and biological activities.

The potentially pathogenic bacterial agents recorded in this study are essentially gram negative enteric bacteria and few other gram positive. These organisms which were also reported earlier on (5,6,7) include *Klebsiella* species, *Escherichia coli*, *proteus* species, *Pseudomonas* species, *Salmonella* species *Staphylococcus aureus*, *Staphylococcus albus* and yeast cells. According to Ashiru and Osoba (23) a number of human diseases have been attributed to have originated from community acquired bacterial agents, especially where environmental conditions such as poor sanitation, heavy flies density and indiscriminate disposal of human and animal waste is prevalent. It is important to note that the heap of refuse dumps in the study are located at a no far distance from the

market centre where arrays of exposed food items are displayed. Earlier on, Adeyeba and Okpala (24) have reported that common filth houseflies are active mechanical transmitters of potentially pathogenic parasites and bacterial agents in Ibadan markets.

The prevalence of the potentially pathogenic bacterial agents in refuse dumps in Ibadan market further confirms the report of Adeyeba and Okpala (24) on enunciated. This result shows a varying degree of multiple bacterial contamination. Though the variation was generally statistically significant ($X^2 = 251.50$, $df=4$, $P<0.001$) in the study area the degree of bacterial contamination in Sang market (15.4%) and Oje market (15.9%) was not significantly different.

Prevalence of these multiple bacterial agents varies with change in climatic condition. It has been shown that at mean air temperature $26.1 \pm 0.5^\circ\text{C}$ and mean relative humidity of $82 \pm 30\%$, more bacterial agents were isolated in the refuse than at mean air temperature $28.4 \pm 0.5^\circ\text{C}$, mean relative humidity of $72 \pm 3.5\%$.

The difference in the prevalence by climatic condition was statistically significant ($X^2 = 834.23$ $df=4$, $P<0.001$). This reinforces the fact that the survival of bacterial agent depends on conducive atmospheric conditions among things as opined by Adeyeba and Okpala (24).

RECOMMENDATION AND

CONCLUSION

This study has shown that there is a high degree of refuse contamination with pathogenic intestinal parasites and bacterial agent in Ibadan market places. These reservoirs of potential infectious agents portends a great danger public health as most food stuffs sold in the markets are often left exposed to

house flies which mechanical carrier of pathogenic in the area (24). It is our considered opinion that the waste dumps in Ibadan could be turned to a useful economic resource as against the present status of "nursery" of pathogens. Therefore the mountainous refuse dumps could be processed into organic fertilizer for use by the farming community of the state to boost the economy of the area. the fertilizer plant would also provide job opportunity for the youth as part of the poverty alleviation programme of government. Besides, the roads would be cleared of the mountainous rubbish to ensure vehicular and pedestrian movement. It is also recommended that the Health Education unit of the Local government should be adequately funded in order to perform its traditional rôle/duty of informing, educating and communicating. This will ensure that the knowledge, attitude and beliefs of the selling and buying population are changed positively to promote good public health.

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CUTANEOUS AND INTESTINAL MYIASES IN LAGELU L.G.A OF OYO STATE.

OLUWATOSIN M.A **, FADAHUNSI I.F* Dept. of Med. Microbiology and Parasitology, College of Health Sciences, LAUTECH Osogbo**, Dept. of Microbiology, University of Ibadan, Ibadan*

**Corresponding Author

Running Title: Incidence of Myiasis in Ibadan suburbs, Oyo State.

Key Words: Fly larve, Myiasis, Pustular eruptions, manual extraction, Itchin Tumbu-fly.

There cases of cutaneous myiasis by *Cordylobia anthropophaga* and a case of intestinal pseudo-myiasis involving *Eristalis specie* are reported in patients from Oyedeji, Apatere and Dagbolu all in Lagelu Local Government area of Oyo State, Nigeria. All cases involved children except the one of multiple cutaneous type which was observed in an adolescent female patient.

Patients' conditions gradually improved after recovery of the larvae from them. This report constitutes the first recorded autaneous and intestinal myiasis in this rural area, albeit many such cases in the past have gone unrecorded.

INTRODUCTION

Myiasis is the infection of living tissues of vertebrate animals (and Man) by dipterous fly larvae. Several genera of calliphorid flies cause myiasis in man and animals in tropical Africa (1).

Human countaneous myiasis has been reported in various parts of the country (2-5). Human intestinal myiasis in Nigeria is not rampant neither is it frequent (6), and many reported cases involved the larvae of the syrphid fly called *Eristalis*. Intestinal and rectal myiasis by *Eristalis* have been reported form south Africa (7), Zambia (8) and Egypt (9). None so far has been reported in Nigeria, although cutaneous myiasis due to *Eristalis luteola* involving breast tissue invasion in a female patient has been described by Ogunba (unpublished work) in Ibadan.

The present report presents three cases of cutaneous myiasis by *C. anthropophaga* and one intestinal form by *Eristalis spp* in patients from Oyedeji, Apatere and Dagbolu, all reporting as outpatients at LGA Clinic based at Oyedeji, in Lagelu Local Government Area of Oyo State.

PATIENTS AND METHODS

Case 1

A 14yr-old female student from Apatere Community Secondary School was first seen at the L.G.A Clinic, Oyedeji in early May, 2001. She looked pale and tired, and complained of intense itching. Multiple pustular eruptions of about one week duration were observed on her trunk, groins and thighs. Single lesions also occurred on her left upper eyelid and neck. When a lesion was squeezed, a small opening (1mm diameter) exuded a serous fluid through which a maggot was gently milked out. In all, 14 larvae were manually extracted over a period of two days. Six were extracted in day one and eight in day-two. Five were extracted from her trunk, three from her pubic region, four from her thighs and one each from her left eyelid and her neck.

The larvae were identified as those of *C. anthropophaga*. Peripheral blood examination of this patient revealed Hb of 11.1g/l, PCV of 36%, WBC of 14.0 x 10⁹ /l, Neutrophils 84%, Lymphocytes 11%, Monocytes 5%. The lesions healed rapidly following extraction of the larvae.

Case 2

A 7yr-old boy brought from Dagbolu village was seen at the LGA Clinic, Oyedeji in late May, 2001. The boy had a solitary enlarged furuncle of about 8 days duration. The lesion which occurred below the left axilla was reddened and tender. The boy was diagnosed as a case of cutaneous myiasis. When the lesion was firmly pressed, it yielded a creamy-white larva of *C. anthropophaga*. On further enquiry from the mother of the boy, she admitted that the family kept a dog but could not recall any incidence involving the animal and any of other members of the household.

After extraction of the larva, healing of the lesion took a short duration of time.

Case 3

A boy of 1½ years of age whose parents reside in Oyedeji was brought to the Clinic in early June, 2001 with a discharging sinus on the back of the left thigh. This had caused the boy excessive itching and restlessness for a period of three days, with attendant crying, loss of appetite and sleepless nights.

The exudates from the sinus was purulent. When gentle pressure was applied around the lesion, a live creamy-white larva of *C. anthropophaga* was extracted.

On further enquiry, the mother disclosed that the family did not keep animal pets, and none of the boy's brethren had any such lesions. The lesion healed within three days after the extraction of the larva.

Case 4

A 9 years old girl was brought from Dagbolu to the LGA Clinic at Oyedeji in early November, 2001. She was pale and constipated, and had abdominal distention. On enquiry,

her mother revealed that the daughter had been treated for fever with a herbal concoction at Dagbolu village. Subsequently the girl developed abdominal pain, nausea, an irritation in her throat which frequently resulted in coughing and vomiting.

While at the Clinic the girl coughed convulsively and vomited two motile creamy-white larva. The larvae had globular anterior regions, and tail-like retractile posterior ends fringed with setae.

The two larvae were indentified as the third instar larvae of *Eristalis specie*.

Stool sample taken from the girl was examined and yielded no maggots (larvae) but showed ova of *Ascaris lumbricoides* and *Trichuris trichiura*. The girl was immediately placed on Combantrin (Pyranterol pamoate) but could not be followed up for further assessment because she defaulted clinic attendance henceforth.

DISCUSSION

In Nigeria, the tumbu fly (otherwise called *Cordylobia anthropophaga*) is an important cause of cutaneous myiasis. Female flies may oviposit on laundry exposed to air-dry in unprotected areas or on dry soils contaminated with human or animal urine or excreta.

Larvae from hatched eggs may temporarily attach to soiled clothes, and eventually penetrate the skin if such clothes are worn without ironing (10).

The distribution of lesions observed in case one was suggestive of infestation acquired from contaminated clothings although the patient did not recall any unusual incident about her clothings.

We noted that, of the three cases of cutaneous myiasis described here, two involved children below 10 years of age. The preponderance of children involvement in myiasis is a common observation (2-5). It was suggested (5) that the increased risk of infestation in children is probably due to frequent contamination of their dresses and

uncovered bodies, sleeping and playing areas with urine, faeces (fresh or dried) and other fly attractants, as well as children's closer contact with pets and other domestic animals.

Most cases of *C. anthropophaga* myiasis have been described from the western (2,4,5) and northern (3) parts of Nigeria. Iwuala and Onyeka (11) have noted that the distribution of *C. anthropophaga* ranges eastwards beyond the river Niger. Furthermore, it has been noted (12) that the fly is endemic in the neighbouring Cameroon Republic. Nevertheless, the authors of this work are not aware of any previous report documenting its occurrence in Lagelu Local Government area of Oyo State Nigeria, most especially in the present rural area being investigated.

Because our patients had not traveled outside the local government for several weeks, we presumed that infestations were acquired locally.

The saprophytic "rat-tailed" maggots of *Eristalis specie* may occasionally cause intestinal or rectal myiasis in man (6-8), and has been incriminated in 36 out of 156 cases of human intestinal infestations (13). Symptoms associated with intestinal invasion include seizures, colicky abdominal pain and bloody stool (13). These symptoms, with the exception of passage of bloody stool, were noted in the patients involved in this report. Not only intestinal myiasis, urinary myiasis involving *Eristalis specie* have been described (14) in a male patient with manifestations of urinary tract infection.

In intestinal myiasis caused by *Eristalis specie* the route of entry is presumed to be oral. Eggs presents in such infected water may initiate intestinal myiasis (10) but most infestations result from ingestion of viable larvae which may remain in the intestines, and may be excreted or vomited later (10,13).

The evidence of herbal treatment as seen in case four in this report, suggested the ingestion of larvae present in a contaminated medication. As with most reports of human intestinal tract for a long period of time, hence a case of accidental or pseudo-myiasis can be presumed.

The authors of this report did not carry out any house-to-house case study to assess or determine the extent of human or animal involvement in these infestations. However, the information volunteered by parents of the patients suggested that our observations only happened to be a recorded case out of many cases that silently went unreported to the clinic at Oyedeji.

However, the observations in this report confirmed the occurrence of myiasis-producing flies in this rural area of the local government although the magnitude and implications of the problems are yet to be fully assessed.

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ANTIBIOTIC SENSITIVITY OF ISOLATES OF *PSEUDOMONAS AERUGINOSA* IN ENUGU, NIGERIA

BY: U.C. OZUMBA, Department of Medical Microbiology
University of Nigeria Teaching Hospital Enugu, Nigeria

The pattern of antibiotic sensitivity of 229 clinical isolates of *Pseudomonas aeruginosa* isolated between June 1998 and May 2000 at the University of Nigeria Teaching Hospital (UNTH) Enugu was studied. The isolates were recovered from various clinical specimens by culturing on standard media viz: blood agar, macConkey agar and Cled agar and identified by routine procedures. Antibiotic sensitivity tests were performed by the disc diffusion technique employing multidisc (habdisc) and using sensitivity test agar incubated at 37°C for 24 hours. The results were read and interpreted according to the manufacturer's instructions. Majority of the isolates tested were susceptible to Ceftazidime (88.5%), Colistin (83.75%), Ciprofloxacin (62.1%) and Ofloxacin (62.5%). Non-urinary isolates were more sensitive than the urinary isolates to ofloxacin, Gentamycin, Streptomycin, Ceftriaxone and Cephtazidime. Similar incidence of resistance was observed between the two groups to other antibiotics. Efforts must be made to improve infection control practises, improve antimicrobial utilization practices and establish an antibiotic policy for the country.

INTRODUCTION

Pseudomonas aeruginosa is an ubiquitous organism and has a world wide distribution (1,2). It is a major problem as a multi resistant nosocomial pathogen(3). Before the 1940s, *Pseudomonas* infections were rare but those organisms have become among the more common opportunists that infect debilitated, burned or immuno suppressed individuals(4). *Pseudomonas aeruginosa* seldom causes infection in healthy people. In hospitals however, it's natural resistance to many antimicrobials and it's ability to grow in solutions used for treatment make it difficult to control.

The present communication reports on the invitro antibiotic sensitivity pattern of all isolates of *Pseudomonas aeruginosa* from diverse clinical specimens between June 1998 and May 2000 in our hospital. The project was divided into two parts.

1. The resistance patterns of the isolates were examined from an overall perspective,

irrespective of the site of isolation.

2. A comparison of the resistance pattern was made between urinary and non-urinary isolates.

MATERIALS AND METHODS

A total of 229 clinical isolates of *pseudomonas aeruginosa* was tested for invitro antibiotic sensitivity to 15 antimicrobial drugs between June 1989 and May 2000 at the university of Nigeria Teaching Hospital (UNTH) Enugu. The specimens were classified into two groups: non-urinary and urinary specimens. The group included wound swabs, blood body fluids, specimens from respiratory tract, ear, nose and throat, genital and conjunctival regions. The isolates were recovered from various clinical specimens by culturing on standard media viz blood agar, macConkey agar and Cled agar. Identification of the isolates was based on characteristic colonial morphology production of pigment pyocyanin, characteristic smell and production of oxidase.

Antibiotic sensitivity tests were performed by the disc diffusion technique employing multidisc (habdisc) and using sensitivity test agar incubated at 37°C for 24 hours. The results were and interpreted according to the instruction of the manufacturer. Due to the nonavailability of some antimicrobial discs at certain periods, not all antimicrobials were tested in equal numbers.

RESULTS

Out of the 229 isolates included in the study, 187 were from adults and 42 from children: 143 from males and from females. One hundred and five (105) isolates were from urine while 124 were from other sources.

1. Overall Pattern

The results of antibiotic sensitivity tests with the isolates are given in (Table I). The number of isolates tested varied depending on the availability of antimicrobial discs. As can be seen majority of the isolates tested were susceptible to Ceftazidime (88.5%), colistin (83.7%), ciprofloxacin (62.1%) and ofloxacin (62.5%). Other antibiotics were effective for a lesser number of isolates.

2. Comparison of Urinary Versus Non-Urinary Isolates

Similar incidence of resistance was observed among urinary and non-urinary isolates of *Pseudomonas aeruginosa* for most of the antibiotics except for Ofloxacin, Gentamycin, Streptomycin, Ceftriaxone and Ceftazidime where non-urinary isolates were more sensitive than the urinary isolates (Table II).

TABLE 1: IN-VITRO ANTIBIOTIC SENSITIVITY OF CLINICAL ISOLATES OF *P. aeruginosa* IN ENUGU, NIGERIA.

Antibiotic	Concentration in the disc	No of isolates sensitive/No. tested	% Sensitive	% Resistance
Ciprofloxacin	5mcg	18/29	62.1	37.9
Colistin	10mcg	200/229	83.7	16.3
Gentamycin	10mcg	105/226	44.1	55.9
Azithromycin		13/29	44.8	55.2
Pefloxacin	5mcg	73/131	55.7	44.3
ceftazidime	30mcg	108/122	88.5	11.5
Ampicillin	10mcg	20/148	0	100
cotrinnoxazole	25mcg	1/150	0.7	99.3
ceftriaxone	30mcg	55/122	45.1	54.9
Tetracycline	30mcg	2/154	1.3	98.7
Nitrofurantoin	50mcg	3/45	6.7	93.3
Nalidixic acid	30mcg	2/46	4.3	95.7
Ofloxacin	5mcg	10/16	62.5	37.5
Streptomycin	10mcg	32/86	37.2	62.8

TABLE II: In-Vitro antibiotic sensitivity of Clinical Isolates of *Pseudomonas aeruginosa* in Enugu, Nigeria, Urinary versus Non-urinary Isolates.

Antibiotic	No of isolates No of tested	Sensitive/ No of tested	% Sensitive	% Resistance
Ciprofloxacin	12/19	(6/10)	63.2(60)	36.8(40)
Colistin	88/110/129	(112/129)	80(86.8)	20(13.2)
Gentamycin	37/103	(68/133)	35.9(51.1)	64.1(48.9)
Azithromycin	4/9	(9/20)	44.9(45)	55.6(55)
Pefloxacin	40/65	(33/66)	61.5(50)	38.5(50)
Ceftazidime	45/53	(63/69)	64.9(91.3)	115.1(8.7)
Ampicillin	0/58	(0/90)	(-)	10.0(100)
cotrimoxazole	0/150	(1/150)	- (0.7)	100(99.3)
Rocephine	26/57	(29/65)	45.6(44.6)	54.4(55.4)
Tetracycline	0/154	(/154)	- (1.3)	100(96.7)
Cefuroxime	3/56	(5/75)	.4(6.7)	94.6(93.3)
Nitrofurantoin	3/5	(-)	4.3(-)	95.7(-)
Nalidixic acid	2/46	(-)	4.3(-)	95.7(-)
Streptomycin	9/35	32/86	25.7(37.2)	74.3(62.8)
Ceftriaxone	8/16	(10/12)	50(83.3)	50(16.7)
Ofloxacin	4/7	(6/9)	57.1(66.7)	42.9(33.3)

Figures in parenthesis represent non-urinary isolates.

DISCUSSION

The resistance of bacteria to antibiotics particularly those used for first-line therapy is an increasing cause for concern (1,5,6,7). In vitro susceptibility testing is crucial to assess the resistance pattern in any specific location and for each individual agent(8). The pattern of resistance may vary in different hospitals, depending on several factors such as antibiotic prescribing policy, types of patients, the level of hygiene and infection control(7). The prevalence of resistance amongst our isolates was very high probably due to very ineffective infection control procedures in the hospital coupled with reliance on a narrow spectrum of antibiotics due to absence of

antipseudomonas antibiotics like amikacin, tobramycin, imipenim, cefsulodin, netilmicine and cefoperazone. The frequency of susceptibility was highest to ceftazidime (88.5%) and colistin (83.7%). The sensitivity of the isolates to Flouroquinolones; Ofloxacin and Ciproflozacin (62.5%) and (62.1%) respectively was much lower than that of others(8). Thus from the present report, it seems that resistance of *Pseudomonas aeruginosa* to the flouroquinolones is on the rise. The antibiotic prescribing policy may be contributory to this, since many physicians in Nigeria, usually prescribe without recourse to antibiotic sensitivity patterns.

Unlike the present study where the sensitivity of isolates to cotrimoxazole, tetracycline and gentamycin was very low (0.7%, 1.3% and 44.1% respectively, others(5), had moderately higher levels of sensitivity (55.17%, 48.28% and 67.75%) respectively. In Nigeria, cotrimoxazole and tetracycline are amongst the four most commonly abused antibiotics. They are easily available in chemists' shops without prescription. Non-urinary isolates were more sensitive to Pefloxacin, Gentamycin, Streptomycin, Ceftriaxone and Ceftazidime than urinary isolates. This may be due to the facts that isolates from the urinary tract were from patients who were catheterized at one period or the other thus predisposing to recurrent infections with more antibiotic resistant organisms.

In view of the occurrence of multiple antibiotic resistant strains, coupled with the increasing resistance of *P. aeruginosa* to the flouroquinolones, considerable effort must be made to establish and improve infection control units, antimicrobial utilization practices and establish an antibiotic policy for the hospital and the country.

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METHICILIN-RESISTANT STAPHYLOCOCCUS AUREUS (MRSA) AT JOS UNIVERSITY TEACHING HOSPITAL.

E.I. IKEH, Department of Medical Microbiology, Faculty of Medical Sciences,
University of Jos, P.M.B 2084 Jos, Nigeria.

KEY WORDS: MRSA, PREVALENCE, SUSCEPTIBILITY, ANTIBIOTICS.

A prospective surveillance of Methicillin resistant staphylococcus aureus (MRSA) was carried out at Jos University Teaching Hospital, Nigeria, over a one year period. This study highlights the continuous importance of MRSA in causing both hospital and to a less extent community acquired infections. Out of the 180 consecutive isolates of *S. aureus* tested, 758 (43%) were found to be methicillin resistant, 81% (63 isolates) of the MRSA were from hospital in-patients while 19% (15 isolates) were from out-patients. The highest rate of methicillin resistance (81%) was found in surgical wound infections while the special care baby unit (SCBU) service recorded 4%. 85% of the MRSA were sensitive to Ofloxacin while 46% were sensitive to peflaxine. Most MRSA isolates were multiply resistant to Augmentin, centriaxone and ceftazidime, thus confirming the nosocomial nature of the isolates. Vancomycin and teicoplanin are not locally available and so ofloxacin is the drug of choice. This study has demonstrated a high prevalence of MRSA in our hospital, which definitely plays a significant role in hospital acquired infections. In conclusion, the relatively high prevalence of MRSA in this study has shown that there is a "limited" level of infection control activity in our hospital.

INTRODUCTION

Hospital infections are all those infections acquired in hospital, which were absent at the admission. A few of the patients become infected with some consequences for them such as complications and prolongation of hospital stay, for their comminutes such as diffusion of infection, and for the hospital as loss of time and resources because of the use of higher quantities of antibiotics. *Staphylococcus aureus* is a ubiquitous organism which is present in the anterior nares of approximately 40% of healthy adults is mainly transmitted by hand contact (1), and is frequently implicated in nosocomial infections. The most remarkable feature of *S. aureus*, however, is its ability to acquire resistance to antibiotics. Many resistance genes are acquired by plasmid-mediated gene transfer, and some may be transferred to the chromosome as mobile genetic elements (2). Probably, the most significant achievement of *S. aureus* has been the acquisition of methicillin resistance. Emergence of MRSA made clinical use of vancomycin inevitable, and

predisposed *S. aureus* towards acquisition of Glycopeptide resistance. MRSA is a generic term for all *S. aureus* strains carrying *Mef. A* gene (3,4) and expressing certain levels of methicillin or oxacillin resistance.

MATERIALS AND METHODS

(a) Study Area

Jos University Teaching Hospital (JUTH) is a 521-bed tertiary hospital with acute, general and specialist units for both paediatric and adult patients. It also serves as a referral hospital.

(b) Method

The susceptibility of consecutive isolates of *S. aureus* (isolated within a 12 month period) to Oxacillin was determined on Mueller- Hinton agar supplemented with 2% NaCl. Plates were inoculated by dipping sterile cotton swabs into the suspension of the overnight growth of the organism prepared to a density of a McFarland No.0.5 standard; expressed excess liquid from the swabs and inoculated the surface of the agar by the spread method. The 1 microgram Oxacillin discs were aseptically placed on the surface of the inoculated plates and incubated aerobically at 35°C for 18-24 hours. The isolates were also similarly

inoculated onto the surfaces of plain Mueller Hinton agar plates and Augmentin (30mcg), Ofloxacin (10mcg), Pefloxacin (5mcg) and Ceftazidime (30mcg) discs were placed and incubated as above. The zones of inhibition were measured and compared with NCCLS (5). The isolates that were resistant to Oxacillin (≤ 10 mm diameter) were termed methicillin Resistant S.aureus (MRSA) (5).

RESULTS

This study describes a 12-month audit of MRSA at JUTH. Out of 180 isolates of S.aureus tested, 78 (43%) were found to be methicillin resistant. The MRSA accounted for 66% (57 isolates) of S.aureus from hospital in-patients and 23% (21 isolates) of those from out-patients. The percentage distribution on each service in the hospital for MRSA are shown in Table 1. Surgical services had the highest prevalence of 46%, while general out-patient department (GOPD), medical, paediatric, Special Care Baby Unit (SCBU) and casualty wards had 15%, 12%, 19%, 4% and 4% respectively ($P < 0.05$).

Table 2 shows the percentage distribution of isolated MRSA from each site of infection. Surgical wound infection recorded the highest with 81% while cutaneous, urinary tract and eye infections recorded 8%, 8% and 4% respectively. ($P < 0.05$).

The susceptibility pattern of the 180 S.aureus isolates to Oxacillin and five other antibiotics is shown in Table 3. 57% of the isolates were sensitive to Oxacillin while 43% were resistant (MRSA). For Augmentin, 40% were sensitive while 92%, 12%, 76% and 10% of the isolates were sensitive to Ofloxacin, Ceftriaxone, pefloxacin and Ceftazidime respectively.

The susceptibility pattern of the 78 MRSA isolates are shown in Table 4. 85% of them were sensitive to Ofloxacin while 46% were sensitive to Pefloxacin. The percentage sensitivities to Augmentin, Ceftriaxone and Ceftazidime were 9, 6 and 7 respectively. The percentage sensitivities for the non-MRSA were as follows:- Augmentin (64), Ofloxacin (100), ceftriaxone (9) and ceftazidime (6). Most MRSA isolates were multiply resistant to Augmentin, Ceftriaxone and Ceftazidime, thus confirming the nosocomial nature of the isolates.

TABLE 1: THE PERCENTAGE DISTRIBUTION ON EACH SERVICE FOR MRSA AT JOS UNIVERSITY TEACHING HOSPITAL, NIGERIA.

Service	GOPD	CAS	MED.	SURG	PAED.	SCBU	TOTAL
Number positive	12	3	9	36	15	3	78
% Positive	15	4	12	46	19	4	43

$$X^2 = 72.54; P < 0.05$$

SYMBOLS

GOPD	=	General Out-patient Department
CAS	=	Casualty
MED	=	Medicine
SURG.	=	Surgery
PAED	=	Paediatrics
SCBU	=	Special Care Baby Unit.

TABLE 2: THE PERCENTAGE DISTRIBUTION OF ISOLATED MRSA FROM EACH SITE OF INFECTION AT JUTH

Isolates	Pathogen	Site %			
		SWI	CUT	UTI	EYE
	MRSA	81	8	8	4
	% Distribution	81	8	8	4
	Number	63	6	6	3

$$X^2 = 164.22; P < 0.05$$

SYMBOLS

- SWI = Surgical Wound Infection
 CUT = Cutaneous
 UTI = Urinary Tract Infection
 EYE = Eye

TABLE 3: PERCENTAGE SENSITIVITIES OF 180 S. aureus ISOLATES TO METHICILLIN AND 5 OTHER ANTIBIOTICS AT JUTH, NIGERIA.

Antibiotic = Methicillin Sensitive Resistant	Augumentin		Ofloxacin		Ceftriaxone		Pefloxacin		Ceftazidime	
	Sensitive	Resistant	Sensitive	Resistant	Sensitive	Resistant	Sensitive	Resistant	Sensitive	Resistant
57 43	40	60	92	8	13	86	76	24	10	90
	(171)		(180)		(123)		(138)		(90)	

$$X^2 = 117.18; P < 0.05$$

Figures in parenthesis indicate the number of isolates tested, whenever these were less than the number isolated.

TABLE 4: PERCENTAGE SENSITIVITIES OF MRSA AND NON-MRSA ISOLATES TO 5 ANTIBIOTICS AT JUTH, NIGERIA.

Antibiotic MRSA, n = 78 Non-MRSA, n = 102	Augumentin		Ofloxacin		Ceftriaxone		Pefloxacin		Ceftazidime	
	Sensitive	Resistant	Sensitive	Resistant	Sensitive	Resistant	Sensitive	Resistant	Sensitive	Resistant
	9	91	85	15	6	94	46	54	7	93
	64	36	100	0	9	91	97	3	6	94
	(69)		(60)		(51)		(138)		(39)	

$$X^2 = 157.69; P < 0.05$$

Figures in parenthesis indicate the number of isolates tested, whenever these were less than the number isolated.

DISCUSSION

The study has highlighted the continuous importance of MRSA in causing both hospital and to a less extent community acquired infections. The relatively high prevalence (43%) of MRSA has shown that there is a "limited" level of infection control activity at JUTH. This must have accounted for the high prevalence of post-operative wound infections earlier reported by Ihezue et al (6) in the hospital. The highest prevalence of 46% in surgical services is attributed to the lack of adequate precautions in the surgical wards especially with respect to wound dressing. The low prevalence of 4% MRSA in SCBU is due to the increased awareness amongst the SCBU staff of the need to prevent nosocomial infection in that unit. It has been found out that control and surveillance of nosocomial infection does not have a priority at senior management levels within the hospital. Thus immediate infection control measures with emphasis on vigilant and careful handwashing before and after patient contact, strict isolation measures, culture surveillance, inter-hospital transfer policies and in-service education will ultimately reduce the incidence of nosocomial MRSA in our hospital. A very effective infection control committee in the hospital will help to co-ordinate the above measures as the costs of an MRSA outbreak both financially and psychologically cannot be overemphasized. In the absence of the glycopeptides (vancomycin and teicoplanin) in Nigeria, Ofloxacin and to a lesser extent pefloxacin are the antibiotics of choice for the treatment of MRSA infections in our local setting. In order to preserve the efficacy of these two drugs the following interventions should be implemented:- (a) prospective identification of patients at risk (risk profiling); (b) institution of proven preventive strategies; (c) rapid identification of infection sources in high risk individuals (d) monitoring the prevalence of antimicrobial resistance for individual pathogens and

(e) appropriate selection of antibiotics and adjunctive therapy.

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SCREENING OF FEBRILE CHILDREN ON HOSPITAL ADMISSION FOR URINARY TRACT INFECTIONS (UTI).

BY: ADEDOYIN O.T ; OYEYEMI B.O; AIYEDEHIN O.V., Department of Paediatrics, University of Ilorin Teaching Hospital, P.M.B. 1459, Ilorin. Corresponding Author: DR. O.T. ADEDOYIN, Department of Paediatrics, University of Ilorin Teaching Hospital, P.M.B. 1459, Ilorin.

Urinary tract infection (UTI) is one of the most often missed diagnosis in children in the tropics. This is because of the varied and similar presentation of UTI to other common illnesses. A total of 154 patients with various presumptive clinical diagnosis at admission were screened for the presence of UTI. Only 33 (21.4%) patients had proven UTI. Majority of these patients (20 or 60.6%) were aged < 5 years. The findings of UTI was more amongst patients with presumptive clinical diagnosis of bacteria infections (like sepsis, typhoid septicaemia, bronchopneumonia etc.), and severe malaria. The commonest organisms isolated were *Escherichia coli* 12(36.4%) and *Klebsiella* 12(36.4%). There was increased sensitivity of these organisms to both ceftazidime and the quinolones. It is concluded that there should be high index of suspicion of UTI in patients with bacteria infection (Localised or generalized) and severe malaria particularly those with black water fever.

INTRODUCTION

One of the most often missed diagnosis in paediatric wards in the tropics is urinary tract infections (UTI). This may be due to the inability of some category of paediatric patients to make complaints referable to the urinary tract. The varying and varied presentation of urinary tract infection particularly in children under 2 years where the clinical presentation of UTI could just be failure to thrive, anorexia, feeding difficulty, irritability and even vomiting may also be contributory (1). There is also the tendency to first suspect that most febrile illnesses in the tropics must be due to malaria. All these reasons may have contributed to making the diagnosis of UTI less common, yet UTI portends grave long-term consequence if not attended to promptly. Studies have shown that UTI whether symptomatic or asymptomatic are of greater significance in childhood than in adults as most renal scars occur after such infections within the first five years of life (2-4). Furthermore about 20% of children with end stage renal disease would have had pyelonephritis earlier on in life(5).

In view of all these, the current study aim to determine the presence or absence of UTI in febrile paediatric admission cases at the University of Ilorin Teaching Hospital.

RESULTS

A total of 154 febrile children were screened for UTI during the study period. This includes 97males and 57females giving a male: female ratio of 1.7:1. The age range of the patient was 2months - 16years. There were a total of 32, 21, 38, 36, 24 and 3 patients in the age group 0-1, 1-2, 2-5, 5-10, 10-15 and > 15years respectively. Majority 20(60.6%) were aged < 5years. More females 20(60.6%) than males 13(39.4%) had UTI. Only 3 patients died indicating 2% mortality. One of the three deaths was in a sickle cell disease patient managed for sepsis, who also has UTI due to coliform organisms.

Out of the 154 patients analysed, only 16 had symptoms referable to the urinary tract out of which 6(37.5%) were positive for UTI. Out of 26 cases of severe malaria screened, 5(19.2%) were positive for UTI with two of them having cerebral malaria, two had blackwater fever and one had severe anaemia

secondary to malaria (Table 2). Out of 40 patients with varied bacterial infection, 11 (27.5%) had proven UTI with majority of these cases, 7 (63.6%) having presumptive diagnosis of sepsis. Furthermore two of them had presumptive diagnosis of typhoid septicaemia including one who underwent operation for typhoid perforation. Three (17.6%) of 17 patients with malignancies had UTI, while only 1 (10%) of the 10 malnourished patients had UTI. That patient was also Human Immunodeficiency Virus (HIV) positive. Out of the 12 patients with renal disorders that developed fever and were screened for UTI, there were three (25%) cases of UTI. (Table 2).

The other cases of UTI occurred in the patients with Guillain-Barre syndrome, (GBS), spina bifida and a child with encephalitis/severe brain damage who was catheterised. The organisms isolated in the GBS and severe brain damage patients were E.coli and Klebsiella respectively, while candida was recovered in the patient with spina bifida (Table 2).

Outcome was not influenced by the presence of UTI as 2 (6.1%) out of 36 patents with UTI died compared to 1 (0.8%) out of 121 patients without UTI ($P < 0.05$) (Table 4).

The commonest organism isolated were *Escherichia coli* (*E.coli*) in 12 (36.4%) patients and *Klebsiella* in 12 (36.4%). Others include coliform organism in 4 (12.1%), *Pseudomonas aeruginosa* 3 (9.1%) *streptococcus faecalis* 1(3.0%), and *candida* species 1(3.0%) (Table 5).

There was 88.9% sensitivity of *Escherichia coli* to ceftazidime, 85.7% sensitivity to ciprofloxacin, and 66.7% sensitivity to gentamicin and 70% sensitivity to ofloxacin.

There was 71.4% sensitivity of klebsiella to ceftazidime, 70% to ofloxacin, 90% to ciprofloxacin and 62.5% to gentamicin. There was 100% sensitivity of coliform to gentamicin, ofloxacin, ciprofloxacin and ceftazidime (Table 6).

TABLES

Table 1

Age ranges of subjects in the study with or without UTI.

Age Range (Years)	Presence of UTI	No UTI	Total	% with UTI
0 - 1	6	26	32	18.8
1- 2	6	15	21	28.6
2 - 5	8	30	38	21.1
5-10	8	28	36	22.2
10-15	5	19	24	20.8
> 15	0	3	3	0
	33	121	154	

Table 2

Clinical diagnosis of patients with and without UTI

Clinical Diagnosis	Presence of UTI	No UTI	Total	% with UTI
UTI	6	10	16	37.5
Severe malaria	5	21	26	19.2
Bacterial infection	11	29	40	27.5
Neurological disorders	3	0	3	100
Malignancies	3	14	17	17.6
Malnutrition	1	9	10	10
Renal disorders	3	9	12	25
Diarrhoeal illness	0	10	10	0
Sickle Cell Disease	1	6	7	14.3
Tetanus	0	10	10	0
Hepatitis	0	3	3	0
	33	121	154	

Table 3

Sex of subjects with and without UTI

Sex	Presence of UTI	No UTI	Total	% with UTI
Male	13	84	97	13.4
Female	20	37	57	35.1
Total	33	121	154	

Table 4

Outcome in subjects with and without UTI

Outcome	Presence of UTI	No UTI	Total	% with UTI
Dead	2	1	3	66.7
Alive	31	120	151	20.5%
Total	33	121	154	

Table 5

Organisms isolated in the subjects with UTI

S/NO	Organisms isolated	No UTI	% of Total
1	<i>Escherichia coli</i>	12	32
2	<i>Klebsiella spp.</i>	12	21
3	Coliform	4	38
4	<i>Pseudomonas aeruginosa</i>	3	36
5	<i>Streptococcus faecalis</i>	1	24
6	<i>Candida spp.</i>	1	3
		33	100%

Table 6

Sensitivity pattern of causative organisms of UTI to drugs tested in percentage (%)

Organisms (%)	Drugs tested								
	Gentamicin	Ofloxacin	Ciprofloxacin	Ceftazidime	Nitrofurantoin	Ampicillin	Tetracycline	Chloramphenicol	Streptomycin
E.coli	66.7	70	85.7	88.9	20	0	0	0	17
Klebsiella	62.5	100	90	71.4	33.3	0	42.9	0	50
Pseudomonas	50	100	100	100	0	-	0	0	0
Coliform	100	100	100	100	0	0	0	0	0
Strept. Faecalis	100	0	0	-	-	0	0	0	100

FIGURES

Figure 1

Clinical diagnosis in the febrile patients used for the study.

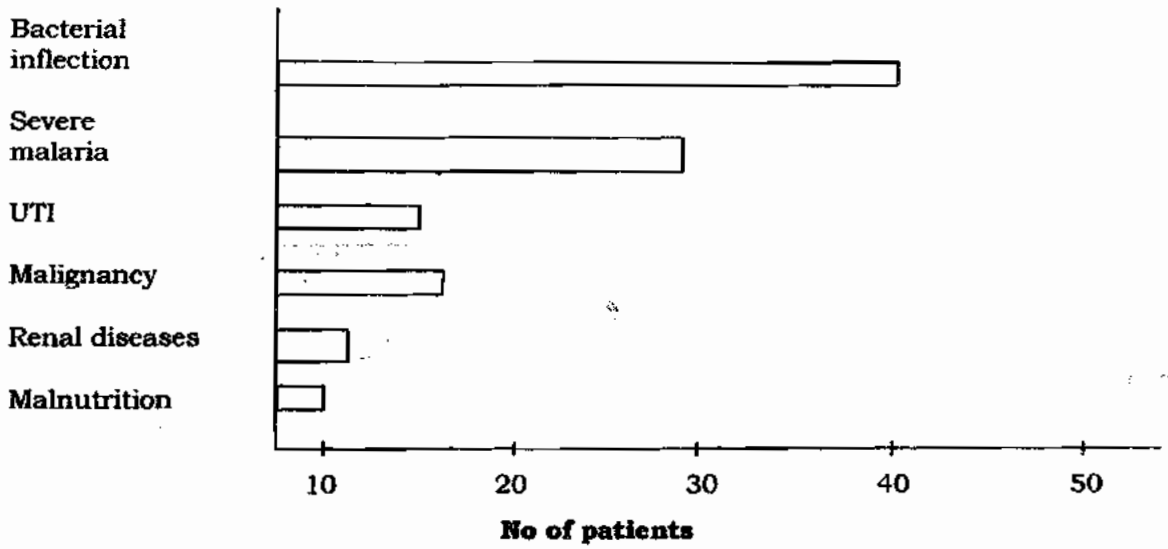
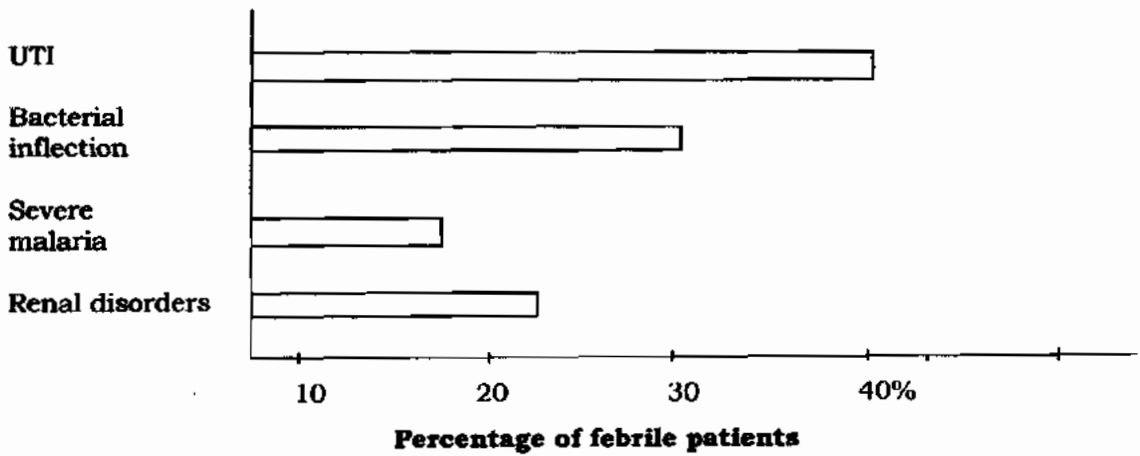


Figure 2

Percentage of febrile patients with the various clinical diagnosis that had UTI



DISCUSSION

The correct incidence of UTI is yet to be obtained in this part of the world as the current study shows that it either just get misdiagnosed as other common childhood illnesses or it could be an associated infection with other illnesses. This trend should be reversed as the management of UTI is usually painstaking in order to identify underlying causes or predisposing factors and to detect complications. Some of these potential complications like secondary vesico - ureteral reflux could lead to pyelonephritis which may produce renal scarring leading to hypertension and possibly end stage renal disease. It has been shown that most renal lesions in adult life may be antedated by untreated asymptomatic bacteriuria in childhood.⁽⁵⁾

The finding of UTI amongst patients with bacterial infection especially sepsis is not surprising. This is because studies have shown that infants with fever, and ill general appearances and those who do not have a potential source of fever such as otitis media e.t.c. on examination have a higher prevalence of UTI.^(6,7) however infants with unequivocal sources of fever on examination such as meningitis etc. are also at risk for UTI but have the lowest prevalence of < 2% ^(6,8). However haematogenous spread of organism causing UTI is commoner in neonates while in older children the mode of spread of the organism is through an ascending infection.⁽⁹⁾ The finding of UTI in patients with typhoid septicaemia is also not surprising since the organism could be seeded in the kidney and subsequently excreted in urine, but it is puzzling that the organism responsible for the UTI is not salmonella typhi but rather *E.coli* and *Klebsiella* in the two cases seen. It may be that the immunosuppression

which may follow typhoid septicaemia makes them susceptible to infection with other organisms (10).

The finding of UTI in patients with malaria indicates that the real cause of the fever may be malaria but there may also exist an underlying asymptomatic UTI because these patients did respond to antimalarial chemotherapy as evidenced by subsidence of the fever. Asymptomatic bacteriuria is very common in children. Workers have reported prevalence of as much as 5% in some studies.⁽¹¹⁻¹³⁾ It may also be that the tubular necrosis resulting from haemoglobinuria as a result of severe malaria predisposed some of them to UTI.

The finding of UTI in patients with Nephrotic syndrome and malnutrition is in agreement with the findings of Ibadin⁽¹⁴⁾ and Ojuawo ⁽¹⁵⁾ respectively. However while the major organisms causing UTI in patients with nephrotic syndrome seen in the Ibadin⁽¹⁴⁾ studies were staphylococcus and untyped coliform. *E.coli* and *klebsiella* were recovered from the two Nephrotic patients with UTI in this study. Furthermore while *Klebsiella* was the predominant organism causing UTI in malnourished patient in the Ojuawo ⁽¹⁵⁾ series, *E.coli* was recovered from the only malnourished patient with UTI in this study.

The finding of UTI in patient with malignancies may be as a result of immunosuppression from the disease or the drugs. But one would have expected some opportunistic organisms being responsible. This was not the case here as *E.coli* and *Pseudomonas* were the offending organisms. The finding of UTI in bedridden patients who were catheterized is also not surprising as prolonged catheterization is capable of resulting in complication of UTI.

The finding of *Escherichia coli* as the commonest organism causing UTI in children is in agreement with findings by other workers ^(12,15). It also indicate that the trend of causative organism of

UTI has not changed. However there was increased sensitivity of the organisms causing UTI to ceftazidime, a third generation cephalosporin and the quinolones. The sensitivity of organisms to ceftazidime agrees with recent findings by other workers (14,15). This is not a cheering news, as the third generation cephalosporins are expensive while the quinolones are contraindicated in children and adolescents in the growth phase. Sensitivity of these organisms to the highly recognized cotrimoxazole was not done but there was reduced sensitivity of these organisms to gentamicin, ampicillin which were once reliable drugs in the treatment of UTI.

It is concluded that there should be a high index of suspicion of UTI in patients with bacteria infection localized or generalized and patients with severe malaria particularly those with blackwater fever. In view of the fact that the sensitivity of these organisms to cotrimoxazole was not tested, it should be noted with caution that ceftazidime and quinolones may be replacing the old reliables.

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NOSOCOMIAL INFECTIONS: URINARY TRACT INFECTION IN PATIENTS WITH INDWELLING URINARY CATHETER.

BY: ** A.A. ONI, G.A. MBAH, M.O. OGUNKUNLE, O. B. SHITTU*, R. A. BAKARE.

Dept. of Medical Microbiology and Parasitology, Dept. of Surgery,
University College Hospital, Ibadan, Nigeria.

** CORRESPONDENCE

With the aim of studying the pattern of urinary tract infection in patients with indwelling urinary catheter in the University College Hospital, Ibadan, a total of 164 patients were recruited. A questionnaire was administered to each patient to provide information on demographic data, clinical diagnosis, and symptoms and signs suggestive of urinary tract infection. Catheter specimen urine from each of the patients was cultured to identify the agents of infection. Antimicrobial sensitivities of the isolates were done. It was found that 54.8% of the patients were above 50 years of age, with a male to female ratio of 2: 1. Benign prostatic hyperplasia was the most common indication for catheterization. 83.5% and 16.5% had Intra-urethral and supra pubic catheterization respectively. 69.5% of these patients had urinary tract infection with 90.40%, 9.6% and 0.9% harbouring 1, 2 and 3 organisms respectively. Intake of antibiotics did not influence the incidence of urinary tract infection. The common agents of infection were *Klebsiella* spp; *Pseudomonas* spp. *Escherichia coli*, *Proteus* spp. *Staphylococcus aureus* and *Candida albicans* in order of frequency. The bacterial agents of infection were resistant to ampicillin, cotrimoxazol and nitrofurantoin commonly used for the patient with urological problem. Ceftazidime, ceftrazone, pefloxacin and ofloxacin showed good sensitivity against the bacteria. These findings should be useful for those who manage patient with indwelling urinary catheter.

INTRODUCTION

Urinary tract infection is an important cause of morbidity and mortality in both adult and children (1). Females are more often affected than males except at the extremes of life (2). Structural abnormalities of the urinary tract make it possible for bacteria that are usually not pathogenic to cause infection (3). Acquired abnormalities such as urinary tract calculi, prostatic hypertrophy, urethral strictures and congenital abnormalities such as double collecting system, and horseshoe kidney all interfere with the free flow of urine and create a complicated setting in which infection is more likely to occur.

Nosocomial urinary tract infections are infections occurring in hospitals and nursing homes. In hospitals where the epidemiology has been better investigated, about 80% of nosocomial urinary tract infections are associated with the use of urinary catheters (4). About 5 - 10% occurs after other genito-urinary manipulations.

The imaginative employment of catheters and drainage tubes in the urinary tract greatly facilitated urological care, however this intubation usually results in bacteriuria (5). Catheters serve as foreign bodies to which bacteria can adhere and prevent the antibacterial function of uroepithelial cell lining the urethra (6). In fact, catheterization of the urinary tract remains the most common cause of nosocomial infection in Medical practice (7).

The microbiology of UTI is predictable with *Escherichia coli*, other Enterobacteriaceae, *Staphylococcus saprophyticus* and *Enterococcus* causing more than 90% of cases. In patients with indwelling urinary catheters bacteriuria is most frequently caused by *Escherichia coli*. Other common organisms are *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Staphylococcus epidermidis*, *Enterococcus* (8), and unusual species such as *Providencia stuartii*⁵. The majority of these bacteria causing urinary catheter associated bacteriuria are from the patient's own colonic flora (9), and may be native

inhabitant or new immigrants, that are exogenous organisms from the hospital environment (10). The endogenous organism may migrate from the perineum to colonise the periurethral area while the exogenous organisms may directly colonize the catheter (11). Organisms may be transferred to the patient by the hands of the health worker. Many of these isolates are resistant to many antimicrobial agents.

In our locality, some workers have documented facts on the microbiology of uncomplicated UTI, but there is little fact on bacteruria and UTI in patients with indwelling urinary catheters. Hence we set out to study the bacterial agents of catheter associated bacteruria and their antimicrobial susceptibility pattern with a view of recommending antimicrobial usage in these patients.

PATIENTS, MATERIALS AND METHODS.

Patients with indwelling urinary catheters in the urological clinic and wards of the University College Hospital (UCH), Ibadan from May to December 1997 were recruited into the study. After verbal consent, a questionnaire was administered to each patient. This gave information on demographic data, clinical diagnosis, indication for, duration and site of catheterization, number and duration of antibiotics taken, and

complaints referable to urinary tract infection.

Catheter urine sample was collected aseptically into sterile disposable screw-capped universal bottle and transported to the laboratory for processing.

Using a standard loop, sample of urine was inoculated onto blood agar and Cysteine-lactose-electrolyte-deficient (CLED) medium. After overnight incubation aerobically at 37°C, organisms were identified to species level by using standard bacteriological techniques (12). The antimicrobial sensitivities were determined by using Stoke's disc diffusion technique (13).

RESULTS

The most prevalent age group with indwelling urinary catheter was 61 -70, being 25.0% of all the patients studied. This was closely followed up by age group 51 -60. Considering all the patients 54.8% were above 50 years of age. The male to female ratio is 2:1.(Table 1)

Of the 164 patients with indwelling catheter, 83 or 50.6% were patients with primary urological problems.

Benign Prostatic Hyperplasia was the most common indication for catheterization. This is followed by Urethral Structure and General Surgical Emergencies.

Urethral catheterization was used in 83.5% of cases while 16.5% of the patients had suprapubic catheterization. (Table 2).

Table 1: Age and Sex Distribution of patients studied.

Age Range (Years)	Males	Females	Total	%
11- 20	3	3	6	3.7
21 - 30	11	7	18	11.0
31 - 40	12	10	22	13.4
41 - 50	13	9	22	13.4
51 - 60	18	7	25	15.2
61 - 70	32	9	41	25.0
71 - 80	14	8	22	13.4
81 - 90	5	1	6	3.7
>90	1	1	2	1.2
Total	109	55	164	100

Table 2: Diagnosis of patients studied and site of catheterization

Diagnosis	Frequency	%	Site of Catheterisation	
			Urethral	Suprapubic
BPH	40	24.5	38	2
Ca Prostate	17	10.4	15	2
Urethral stricture	21	12.8	3	18
Paraplegia	11	6.7	11	0
Ca Bladder	5	3.1	5	0
RTA	10	6.1	10	0
CVA	14	8.6	14	0
Septicaemia	2	1.2	2	0
Burns	2	1.2	2	0
Renal Failure	5	3.1	5	0
Gen. Surg. Emm.	20	12.3	18	2
Diabetes Crisis	9	5.5	9	0
VVF	2	1.2	1	1
Epilepsy	2	1.2	2	0
Rupture of urethra	2	1.2	0	2
Senile Dementia	2	1.2	2	0
Total	164	100	137	27
%			83.5	16.5

KEY: BPH= Benign Prostatic hyperplasia
 RTA= Road Traffic Accident
 Gen. Surg. Emm. = General Surgical Emergencies
 CVA = Cerebrovascular Accident
 VVF = Vesico - vaginal fistula

Of the 164 patients, 84 or 51.2% were on antibiotics either prophylactically or therapeutically, 72(43.9%) had one type of antibiotics 10(6.1%), and 2(1.2%) had a combination of two or three antibiotics respectively, while 80 (48.8%) had none. Considering patients with antibiotics, 26(30.95%) had no organism in urine; 58 had bacteriuria. Of these, 51 (87.93%), 6(10.34%), and 1(1.72%) had 1, 2, and 3 organisms respectively. Of the 80 patients without antibiotics, 25(31.25%) had no organism in urine 55(68.75%) had bacterium. Of these,

50(90.9%) had one organism while 5 (9.1%) had two. (Table III). There is no significant difference between the group with antibiotic and those without antibiotics (p value = 0.85497196).

Of these patients, 122 (73.4%) had no complaints referable to UTI during the period of catheterization 37 (27.0%) of patients with urethral catheter and 5 (18.5%) of those with suprapubic catheter showed features of urinary tract infection. 7 (5.1%) and 4 (2.9%) of patients with urethral catheter had frank discharge and pain around catheter respectively. 3.7% of the patients had suprapubic pain. (Table 4).

TABLE 3: ANTIBIOTIC INTAKE AND FREQUENCY OF ISOLATION OF ORGANISMS FROM URINE.

PRESENCE OF ISOLATES	INTAKE OF ANTIBIOTICS		
	YES	NIL	TOTAL
NO ISOLATE PRESENT	26	25	51
ISOLATES PRESENT	58	55	113
TOTAL	84	80	164

P>0.5

TABLE 4: Complaints referable to UTI and site of Catheterization.

Site	Discharge	Pain around catheter	Suprapubic pain	Fever	Others	No Complaints	Total	%
Urethral	7	4	5	13	8	100	137	83.5
Suprapubic	0	0	1	2	2	22	27	16.5
Total	7	4	6	15	10	122	164	100
%	4.3	2.4	3.7	9.1	6.1	73.4	100	

Of these patients, 50 (30.5%) had no organisms in their urine, while 114 (69.5%) had significant bacteriuria. Of these, 103 (90.4%) had one organism, while 11 (9.6%) were polymicrobial with 10 (8.8%) and 1 (0.9%) having combination of two and three agents respectively.

Table 5 shows the agents of infection in the patients.

A total of 126 organisms were isolated from the urine of these patients. *Klebsiella* species was the most common organism being 36.5% of the isolates. This is followed by *Pseudomonas* spp. (25.1%), *Escherichia coli* (23.0%), *Proteus* spp. (10.3%), *Staphylococcus aureus* (4%) and *Candida albicans* (3.2%).

Table 6 shows that majority of the isolates were from patients with primary urological problems.

Table 7 shows the susceptibility pattern of the isolates to the antibiotics used. All the isolates were susceptible to the fluoroquinolones (ofloxacin and pefloxacin). Most of the isolates were susceptible to Ceftriaxone and Ceftaxidime. The commonly used antibiotics, Ampicillin, Cotrimoxazole, Nitrofurantoin and Nalidixic acid showed low effectiveness. All isolates were resistant to Ampicillin and Cotrimoxazole. Gentamicin which was commonly used against *Pseudomonas aeruginosa*, showed unacceptably reduced effectiveness (33.3%). Ofloxacin and Pefloxacin showed effectiveness against all the isolates.

TABLE 5: ISOLATES FROM URINE SPECIMENS OF THE PATIENTS

Isolates	Frequency 1 st Isolates	Frequency 2 nd Isolate	Frequency 3 rd Isolate	Total	%
<i>Klebsiella</i> spp.	42	2		46	36.5
<i>Pseudomonas aeruginosa</i>	17	2		19	15.1
<i>Pseudomonas</i> spp.	11	1	1	13	10.3
<i>Escherichia coli</i>	28	2		29	23.0
<i>Proteus mirabilis</i>	7	2		9	7.1
<i>Proteus</i> spp.	4			4	3.2
<i>Staphylococcus aureus</i>	4	1		5	4.0
<i>Candida albicans</i>	1			1	0.8
Total	114	10	1	126	100

TABLE 6: DIAGNOSIS AND ISOLATES FROM PATIENTS WITH INDWELLING URINARY CATHETER IN IBADAN.

Diagnosis	Kleb. Spp	Pseudo aerug	Pseudo spp.	Esch. coli	Proteus mirabilis	Proteus Spp.	Staph. aureus	Candida albicans	No. of Isolates	%
BPH	14	5	1	8	5	1	0	0	34	27.0
Ca Prostate	5	1	1	8	1	0	0	0	16	12.7
Urethral stricture	6	3	2	3	2	1	2	0	19	15.0
Paraplegia	4	0	0	1	0	0	0	0	5	4.0
Ca. Bladder	1	1	1	1	1	0	0	0	5	4.0
RTA	1	0	3	3	0	0	1	1	9	7.8
CVA	2	2	1	0	0	0	0	0	5	4.0
Septicaemia	1	0	0	0	0	1	0	0	2	1.6
Burns	2	1	0	1	0	0	0	0	4	3.2
Renal Failure	2	1	0	1	0	1	0	0	5	4.0
G.S.E	5	2	3	1	0	0	2	0	13	10.3
Diabetes	1	2	0	1	0	0	0	0	4	3.2
VVF	0	0	0	1	0	0	0	0	1	0.8
Epilepsy	1	0	0	0	0	0	0	0	1	0.8
Rupture	0	1	0	0	0	0	0	0	1	0.8
Senile D	1	0	1	0	0	0	0	0	2	1.6
Total	46	19	13	29	9	4	5	1	126	100

KEY: BPH = Benign prostatic hyperplasia; RTA = Road traffic accident;
 CVA = Cerebro-vascular accident; G.S.E. = General surgical emergencies.
 VVF = Vesico-vaginal fistula; Rupture = Rupture of urethra; Senile D = Senile dementia.

Table 7: Percentage susceptibility of isolates to the antibiotics tested

Isolates	AMP	COT	NIT	NAL	GEN	CAZ	CRO	PEF	OFX
Klebsiella spp.	0	0	33.3	46	37.5	92	70.8	96	100
Pseudomonas aeruginosa	0	0	0	19	33.3	87.4	25	94	100
Pseudomonas spp.	0	0	44.4	39.0	40	94	50	100	100
Escherichia coli	0	0	0	22.2	50.0	89.0	33.3	89.0	100
Proteus Spp.	0	0	0	22.2	50.0	89.0	33.3	89.0	100

KEY: AMP = Ampicillin
 COT = Cotrimoxazole
 NIT = Nitrofurantoin
 GEN = Gentamicin
 NAL = Nalidixic acid
 CRO = Ceftriaxone
 CAZ = Cefazidime
 PEF = Pefloxacin
 OFX = Ofloxacin

DISCUSSION

Urinary tract instrumentation and catheterization are necessary in the care of many patients who are hospitalized or reside on extended care facilities with problems in their urinary system which result to urine retention or incontinence. Unfortunately, urinary tract instrumentation and catheterization can lead to significant morbidity and mortality (14).

The age distribution of patients with indwelling urinary catheter showed 61-70 years age group were more prone to catheter associated bacteriuria. This is because the elderly are more prone to acquired structural abnormalities and neurogenic bladder secondary to stroke or autonomic neuropathy of diabetes than the young people.

In this study, catheter associated bacteriuria was found to be commoner in males than females. Obstruction caused by BPH, Ca Prostate, and Urethral structure is peculiar to the males.

In this centre 51.2% of patients with indwelling urinary catheter were on antibiotics either prophylactically or therapeutically, with 16.7% of them having varying numbers of combination of antibiotics. However the use of antibiotics did not influence the incidence of UTI in patients with indwelling urinary catheter since no significant difference was found between the group that was on antibiotics and that was not ($P > 0.5$).

It is inferred that a lot of wastage of fund goes into the purchase of antibiotics for prophylactic uses for patients with indwelling urinary catheter. Therefore efforts should be geared towards standard catheter care rather than the use of antibiotics unless when indicated for therapeutic purposes.

In this study, Gram negative organisms predominated accounting for 96.0% of all isolates. This is in consonance with the reports of Akinkugbe *et al* (2) and of Ekweozor and Onyemenem in uncomplicated UTI in the same centre (15). The agents were *Klebsiella* species, *Escherichia coli*, *Pseudomonas aeruginosa*, *Proteus* species. *Staphylococcus aureus* was the only Gram-positive cocci found. Other such as *Enterococcus faecalis* and *Staphylococcus saprophyticus* were not encountered. These organisms are more associated with uncomplicated UTI. The presence of *Staphylococcus aureus* is important as it is commonly associated with Nosocomial infections (6,17), the setting under which these patients were taken care of.

Most of the isolates were multiresistant antibiotics commonly used as prophylaxis or treatment in these patient were virtually inactive. These are Ampicillin, Cotrimoxazole, Nitrofuradantion and Gentamicin. These drugs were given routinely after change of Catheters thereby potentiating the emergence of resistant strain over time. Onyemenem and Ekweozor recommended Gentamicin administration to patients severely infected with *Pseudomonas*, *Klebsiella* or *Enterobacter* (15). The low effectiveness of Gentamicin in this study as well as that reported by Oni *et al* (18) counteracts this advise.

This low effectiveness may be due the fact that Cations (eg. Ca^{2+} and Mg^{2+}) present in urine inhibit the activity of Gentamicin and the development of resistance in hospital environment (19) or that the drug is gradually included among the antibiotics prone to abuse.

Most of the Gram-negative bacilli especially *Klebsiella* species and *Pseudomonas* species are intrinsically resistant to most antibiotics, a situation which favours their continued existence in hospital environment (16). This fact greatly contributes to the high incidence of these agents in this category of patients. There is acceptable levels of

sensitivity of the bacteria to Ofloxacin, Pefloxacin, Cefazidime and Ceftriazone.

A lot of caution is required in the use of these new generation antibiotics. This is because there is emergence of resistance to the quinolones as reported in - vitro tests in our environment(18).

Emergence of resistance to the quinolones has also been reported in patients with complicated urinary infection with *Pseudomonas aeruginosa* (20).

These findings should be useful for those who have to manage patients with catheter associated bacteriuria with limited or without laboratory facilities especially in cases where there have been poor clinical response to therapy inspite of adequate use of commonly prescribed antimicrobials".

The association between bacteriuria and subsequent pyelonephritis is well documented in both clinical and laboratory settings. The role of indwelling catheters as risk factors associated with pyelonephritis is also well accepted (21).

Since urinary catheterization is associated with significant morbidity and even mortality the indication for catheterization must be sound and aseptic techniques used. A closed drainage system is required with administration of oral or system antimicrobial agents as at when due should reduce infection.

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THE RELATIVE ROLE OF SERUM ALBUMIN AND URINARY CREATININE AS BIOCHEMICAL INDICES FOR NIGERIANS WITH PULMONARY TUBERCULOSIS.

* S. A. ADEBISI; ** P. O. OLUBOYO and *** O. O. OLADIPO

* Department of Chemical Pathology and Immunology ** Department of Medicine, University of Ilorin, P.M.B 1515, Ilorin, Nigeria. *** Department of Clinical Pathology, College of Medicine, University of Lagos, Lagos

Key words: Nutritional index, weight, BMI, Albumin, Pulmonary tuberculosis.

Running title – Serum Albumin and Nutrition in tuberculosis.

Correspondence to: DR S.A. ADEBISI

Objective: The objective is to evaluate roles of urinary creatinine and serum albumin as biochemical markers for monitoring the nutritional status of pulmonary tuberculosis patients during treatment. **Design:** This was a longitudinal study. Each patient was studied for six months. **Settings:** This study was carried out at University of Ilorin Teaching Hospital. **Subjects:** Forty-five newly diagnosed patients with pulmonary tuberculosis were used for the study. **Intervention (Method):** Forty-five newly diagnosed pulmonary tuberculosis patients were placed on six months short course regimen. Their weight, Body mass index, serum albumin and 24-hour urinary creatinine were determined before treatment, at the end of the 1st, 2nd, 4th and 6th month of treatment. Using ANOVA, the mean values of the weight, BIM and serum albumin were analysed with further analysis paired student T- test of the pre-treatment values with end of 6th month values. **Main outcome measured:** Their weight, body mass index, serum Albumin and 24-hour urinary creatinine were determined. **Results:** Thirty-one patients with mean age of 36.8 years completed the study. The pretreatment mean weight, body mass index, serum albumin were 49.53kg, 17.72kg/m² and 26.7g/L respectively. The corresponding values at the end of the sixth month of treatment were 57.03kg, 20.4kgm² and 39.97g/L. These three variables showed significant upward improvements. **Conclusion:** Both the body mass index and serum Albumin pretreatment values showed that the patients were malnourished at presentation. Serum albumin being more sensitive and more reliable than both weight and body mass index as revealed by this recommended as index for nutritional assessment in patient with tuberculosis.

INTRODUCTION

There is an established association between pulmonary tuberculosis and malnutrition. Morbidity is over three times greater for underweight patients than for overweight ones. Health education on nutrition is therefore an integral part of the management of patients with pulmonary tuberculosis.¹

Compliance with this nutritional advice is usually monitored by weighing the patient during their clinic visits.

Weighing machines need frequent recalibration if the result obtained will be reliable. Bouts of diarrhoea or vomiting after taking the oral drugs will reduce the reliability of the weight in assessing the nutritional status of the patients. The value of plasma prealbumin as a biochemical parameter for monitoring nutrition in

pulmonary tuberculosis has been established⁽²⁾. However the high cost and the non-availability of the assay in most medical centres in the developing countries where pulmonary tuberculosis is prevalent put a serious limitation on the use of plasma prealbumin as an alternative to simple weighing.

Albumin has a half-life of 21 days. It is a good index of body nutrition, more so for patients with pulmonary tuberculosis that have an average of monthly visits to their clinics. Creatinine is produced from creatine and phosphate in skeletal muscle, and is excreted in the urine. Urine creatinine excretion depends on muscle mass and it has been estimated that 1g (9mmol) of creatinine is derived from 17kg of skeletal muscle.⁽³⁾ Twenty-four hour urinary creatinine excretion correlated well with total body muscle mass.⁽⁴⁾

Urinary excretion of creatinine is a recognized biochemical index for laboratory assessment of nutrition.⁽⁵⁾

This study therefore was carried out to determine the degree of malnutrition amongst patients with pulmonary tuberculosis using their body mass index and to evaluate the value of urinary creatinine and serum albumin as biochemical markers for monitoring the nutritional status of pulmonary tuberculosis patients during treatment.

MATERIALS AND METHOD.

All patients were newly diagnosed pulmonary tuberculosis patients from the chest clinic of the University of Ilorin Teaching Hospital. Patients that were included in the study were sputum positive on direct smear by Zhiel Nelson stain: a supportive chest x-ray finding was also mandatory. Those with human immunodeficiency virus infection, evidence of carcinoma or any protein losing disease were excluded. These patients had six months, short course anti-tuberculous drug regimen. This consists of Isoniazid, Rifampicin, Ethambutol and Pyrazinamide. Pyrazinamide and Ethambutol were used only for the first two months of the therapy.

Each individual was weighed wearing light clothing only and without shoes, on a UNICEF Beam Balance scale. Height was measured using the vertical attachment to the scale keeping the head so that the line from the external auditory meatus to the lateral angle of the eye was horizontal. Quetelet's index was calculated from height and body weight measurements. 24-hours

urine collection was done to determine 24-hours creatinine excretion. Blood (10ml) was drawn from one of the antecubital veins, with minimum stasis. Serum was separated by centrifugation after the sample had fully retracted. Where analysis was not possible on a same-day basis, the serum was preserved deep-frozen at -20°C till the following day.

This study was a longitudinal one. Each patient was followed up for a period of six months that the treatment lasted. All indices, urine and blood samples were taken on the first day of visit before commencement of therapy. Subsequent samples were collected from the patients at the end of the 1st, 2nd, 4th and 6th, months of therapy. Drug compliance and effectiveness of therapy was assessed by a combination of pill counting, urine colour changes with rifampicin, and examination of sputum for acid fast bacilli on each visit.

Serum Albumin was estimated by the bromocresol dye-binding technique.⁽⁶⁾ Creatinine was determined using Reberly Folin method based on jaffe's reaction.⁽⁶⁾

Statistical analyses were carried out in an IBM-compatible personal computer using SPSS software. A one-way ANOVA was done for mean values of body weight, body mass index (BMI), serum albumin and 24-hour urinary creatinine excretion: with further evaluation using the paired student t-test. Association between body weight and BMI on one hand and biochemical indices of serum albumin and 24-hour urinary creatinine excretion was evaluated by analysis of the corresponding paired T-test results.

RESULTS

45 patients started the study, 14 of them were lost to follow up; while only

31 completed the study. The 31 patients were made up of 17 males and 14 females with a mean age of 36.8 years. These patients by using their pretreatment data served as their own controls against which their subsequent data were compared.

Table I shows the mean values of their weight, body mass index (BMI), albumin (ALB) and 24-hour urinary creatinine excretion (UCRE). The mean weight before treatment, end of 1st month, end of 2nd month, end of 4th month and at the end of 6th month was 49.53kg, 51.32kg, 52.81kg, 55.27kg and 57.03kg respectively. The corresponding values for BMI was 17.72kg/m², 18.37kg/m², 18.91kg/m², 19.79kg/m² and 20.46kg/m². The mean serum albumin levels before treatment; end of 2nd month, end of 4th month and end of 6th month was 26.71g/L, 31.00 g/L, 34.61 g/L, 37.45 g/L, and 39.97 g/L, respectively. The corresponding values for 24-hour urinary creatinine excretion was 9322mmol, 8910mmol, 8914mmol and 9397mmol. Both the BMI and serum albumin pretreatment values show clearly that these patients were malnourished at presentation.

A one-way analysis of variance was used to compare the various means obtained for each variable.

Table II shows that the F-probability for weight, BMI, Albumin and 24-hour urinary creatinine excretion was 0.031, 0.002, 0.001 and 0.904 respectively. These show that with the exception of the 24-hour urinary creatinine excretion the other three variables of weight, BMI and serum albumin had statistically significant changes amongst their mean values.

Table III shows the result of further analysis with paired sample T-test, using the pretreatment and post treatment (end of 6th month) value. The critical ratio calculated (t-value) was - 7.450, - 7.333, - 11.691 and 0.15 for weight, BMI, albumin and urinary creatinine respectively, while their corresponding P - values were 0.0001, 0.0001, 0.0001 and 0.882 respectively. The correlation coefficient for each pair was 0.846, 0.763, 0.756 and 0.432 respectively. This shows that there were significant differences in the mean pretreatment values of weight, BMI and albumin when compared with their end of treatment mean values.

TABLE I**MEAN VALUES OF WT, BMI, ALB and UCRE**

	Pretreatment	End of 1 st Month	End of 2 nd Month	End of 4 th Month	End of 6 th Month
WEIGHT (WT) kg	49.53	51.32	52.81	55.27	57.03
BODY MASS INDEX (BMI) kg/m ²	17.72	18.37	18.91	19.79	20.46
ALBUMIN (ALB) g/L	26.71	31.00	34.61	37.45	39.97
24 HOUR URINARY CREATININE EXCRETION mmol/24hrs (UCRE)	9322	8910	8752	8914	9397

TABLE II**ANOVA RESULTS OF WT, BMI, ALB, AND UCRE**

Sources of variation	Degree of freedom	Sum of square	Mean square	Variance Ratio	F. Probability
WEIGHT					
Time	4	1119.7	279.9	2.74	0.031
Residual	150	15352.0	102.3		
Total	154	16471.7			
BODY MASS INDEX					
Time	4	149.273	37.318	4.42	0.002
Residual	150	1265.792	8.439		
Total	154	1415.065			
ALBUMIN					
Time	4	3411.20	852.80	10.29	.0001
Residual	150	12432.39	82.88		
Total	154	15843.59			
URINARY CREATININE EXCRETION					
Time	4	9.935E+06	2.484E+06	0.26	0.904
Residual	150	1.437E+09	9.580E+06		
Total	154	1.447E+09			

TABLE III

**PAIRED T-TEST OF PRETREATMENT AND END OF 6TH MONTH
(POST-TREAT)
WT, BMI, ALB, AND UCRE VALUES'**

	t	df	Sig. (2-tailed)	Correlation
Weight	-7.450	30	<0.0001	.846
Body mass index	-7.333	30	<0.0001	.763
Albumin	-11.691	30	<0.0001	.756
24 Hour Urinary Creatinine	0.15	30	0.882	.432

DISCUSSION

A clear association between BMI and incidence of tuberculosis has been established (1,7,8). Tverdal in his work also observed that mortality from tuberculosis showed a decreasing tendency with increasing BMI. This study with a mean BMI of 17.72kg/m² for the patients at presentation confirms the high prevalence of undernutrition amongst patients with pulmonary tuberculosis. Table one shows a steady improvement in the weight, BMI and serum albumin values of the patients as the treatment progresses. The consistent upward trend of these makers of nutritional status shows that they may be reliable parameters for monitoring tuberculosis.

Subjecting the mean values of the patient's weight, BMI, Serum Albumin and 24-hour urinary creatinine excretion to ANOVA give an F-probability of 0.031, 0.002, 0.001 and 0.904 respectively. These values show that there existed statistically significant differences in the mean values of these patients' weight, BMI and albumin as treatment progresses. This is in agreement with the earlier deduction from the simple observation of the trend in these variables.

A closer look at the calculated critical variance Ratio of 2.74 for weight, 4.42 for BMI and 10.29 for serum albumin shows that serum albumin is the most sensitive of the three variables thereby giving it an edge over both the weight and BMI as indices for assessing nutritional improvement in patients with

pulmonary tuberculosis. The above observations and deductions do not hold for 24-hour urinary creatinine excretion as it fails to change significantly (P-value = 0.904). This observation could be due to a number of reasons: First, the changes in renal dynamics as a result of the drug induced hyperuricaemia associated with anti-tuberculosis drug medication. It could also be as a result of the difficulty associated with the collection of 24-hour urine samples.

Further evaluation with paired samples T-test statistical analysis involving the pre-treatment value and the post treatment (end of 6th month) values gave t-values of 7.450, 7.333, 11.691 and 0.15 for weight, BMI, serum albumin and 24-hour urinary creatinine respectively. These values further confirms that the weight, BMI and serum albumin had changed significantly, the serum albumin with t-value of 11.691 is the most sensitive while the 24-hour urinary creatinine excretion t-value of 0.15 which is below the 95 percentile value of 2.042 at 30 degree of freedom on a t-distribution table is statistically not significant.

We therefore conclude that weight, BMI and serum albumin are very reliable markers of nutritional status in patients with pulmonary tuberculosis. Of these three variables serum albumin is the most sensitive. Since serum albumin is free from some of the shortcomings of weight and BMI like the influence of drug induced vomiting and or diarrhoea on weight and BMI, we are recommending this relatively easily available Biochemical marker as a replacement for simple

weighing and calculated BMI in monitoring the nutritional status of patients with pulmonary tuberculosis.

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