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# BACTERIAL ISOLATES ASSOCIATED WITH ORTHOPAEDIC WOUNDS IN TWO MEDICAL CENTRES IN SOUTHEAST NIGERIA

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**Abstract**

Infection of wounds by microorganisms is most often associated with prolonged hospital stay with the attendant risk of acquisition of multiple resistant organisms from medical devices and hospital environment. A total of 520 male and female respondents aged 18 to 50 years were selected for the study from out – patients and in – patients of the two hospitals, between September – June, 2011. The wound swab was collected from 404 (77.7%) respondents, while 52 (12.9%) of the respondents had concomitant infections of 2 bacterial species each and 352 (87.1%) had single bacterial infection each. Five species of bacteria: *Staphylococcus aureus, Streptococcus pyogenes, Pseudomonas aeruginosa, Proteus mirabilis* and *Escherichia coli were* isolated from the patients. A total of 456 bacterial isolates were obtained. The most prevalent isolate (32.7%) is *Pseudomonas aeroginosa* and the least prevalent bacteria (12.9%) are *Streptococcus pyogenes.* The antibiotic susceptibility of the test isolates showed that ciprofloxacin (ciproxin) exhibited the highest growth inhibitory effect on *Pseudomonas aeruginosa, Staphylococcus aureus, Streptococcus pyogenes, Proteus mirabilis* and *Escherichia coli.* Nitrofurantoin had no growth inhibitory effect on the test isolates, except one strain of *Escherichia coli.* The study has shown that bacteria involved in contamination of orthopaedic wounds are treatable with conventional antibiotics available in South-east Nigerian.

Key words: Orthopedic wounds, Bacteria,Antibiotics,Infections.

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## INTRODUCTION

A wound is a breach in the skin and exposure of subcutaneous tissue following loss of skin integrity. A wound provides a moist, warm, nutritive environment conducive to microbial colonization and proliferation (2).

Orthopedic wounds are wounds that result from the disruption of the skin by the bones or joints of the skeleton, for example in fracture or as a result of surgery to correct bone malformation or to remedy a fracture (7).

Despite technological advances in surgery and wound management, wound infection has been regarded as the most common nosocomial infection especially in patients undergoing surgery and its management practices becomes more resource demanding. (2). Although microorganisms are responsible for wound infection, widespread controversy still exists regarding the exact mechanisms by which they cause infection and also their significance in nonhealing wounds that do not exhibit clinical signs of infection (2).

Wound infection could result in prolonged hospital stay, increased trauma care, and high treatment costs if not properly attended to (9). Members of the *Enterobactericeae* family are fast emerging as important agents for the spread of ESBL genes and strains of *K. pneumoniae, K. oxytoca*, and *E. coli* have been reported. Extended-spectrum beta-lactamases (ESBL) are β-lactamases capable of conferring bacterial resistance to the penicillins, first, second, and 3GCs, and aztreonam (but not the cephamycins or carbapenems) and are usually encoded on plasmids which frequently carry genes encoding resistance to other classes of antibiotics (9).

Orthopaedic wound infections results from microbial invasion of severe cutaneous lesion. According to (5), a wound is said to be infected if it contains pus visible to the naked eye whether or not organisms could be cultured from the purulent material. They cause significant morbidity in terms of patients suffering and disability (8). Wound infection is the most frequent cause of hospitalization. Gram-negative bacteria have been reported to be the major etiologies of wound infections (9).

According to Cruse,(5), all open wound containing bacteria and fungi, remain contaminated with varying numbers and species until after the wound has been successfully closed. A lot of microorganisms especially bacteria and fungi are associated with orthopaedic wounds or among orthopaedic patients. No wound is ever closed if it is suspected to be infected. (14).

Radiation therapy patients as well as diabetics, the obese and those that have had a stroke or some sort of peripheral vascular disease are also more likely to develop some sort of wound infection. (5).

Post operative infections such as periprosthetic septic arthritis, post operative Osteomyelitis and deep wound infections are a particularly devastating complication of orthopaedic surgery (14). An analysis of postsurgical wound infections following head and neck surgery demonstrated an increase in the average hospitalization period from 14 days when wounds healed without complication to 24 days when the wounds became infected (2). The invading microorganisms like *Staphylococcus aureus*, *Clostridium perfringes*, *Clostridium tetani*, *Pseudomonas aeruginosa*, and *Vibrio vulnificus* may produce toxins and other substances that increase their ability to penetrate host tissues, thus producing damage within the host and being able to survive despite host defense mechanism (10). In orthopaedics wound, the surgical site infection after implant surgery is a disaster both for the patient and surgeon. This may lead to increased antibiotic use, prolonged hospital stay; prolong rehabilitation, morbidity and mortality (6). Infection of wounds by microorganisms is most often associated with prolonged hospital stay with the attendant risk of acquisition of multiple resistant organisms from medical devices and hospital environment (9).

Thus the investigation is aimed at identifying the causes of the wounds found on these patients after accidental injuries relating to orthopedic wounds, whether male or female and the possible bacterial agents contaminating the wound due to their occupation or environmental conditions, in other to achieve an effective wound treatment and cure to these organisms using proper antibiotics in which these organisms are susceptible to in this part of Nigeria.

## MATERIALS AND METHODS

### Study area and Study population

This study was carried out at 2 tertiary health institutions in Imo State: Federal Medical Centre Owerri and Imo State University Teaching Hospital, Orlu. Imo State situates in Southeastern Nigeria. The state lies between latitudes $5^{O}$ 30′ and $6^{O}$ 15′ North, longitude $6^{O}$ 38′ and $7^{O}$ 18′ East. A total of 520 male and female respondents aged 18 to 50 years were selected for this study. The respondents were selected from out – patients and in – patients at Imo State University Teaching Hospital Orlu and Federal Medical Centre Owerri. The questionnaires were administered by person – to - person contact after explaining the objectives of the study to each respondent and obtaining his willingness to participate in the study.

### Sample collection and administration of questionaires.

The sample collection was between the period of 10 months, from 13th September to 24th June 2011. Each collected sample was labeled with the respondent's index number indicated on the research questionnaire. The questionnaires were administered by person – to - person contact after explaining the objectives of the study to each respondent and obtaining his willingness to participate in the study.

A sterile Evepon swab stick was used to collect wound swab from each patient in the morning hours prior to dressing of the wound in each hospital. The collected samples were transported to the Laboratory immediately by the field assistants and analysed within 30 minutes in the Microbiology Laboratory. The materials used were sterilized by standard laboratory methods (4).

### Isolation of test organisms

The test isolates were obtained by culture method. The wound swabs collected from respondents were inoculated on duplicate plates of Nutrient agar, Blood agar, Chocolate agar, MacConkey (oxoid,India).

The inoculated Nutrient agar, MacConkey agar, and one set of inoculated Blood agar plates were incubated at $37^{O}$C for 24 hours. One set of inoculated Blood agar plates were incubated anaerobically

using anaerobic gas spark jar at 37$^{O}$C for 24 hours. The plates were examined for microbial growth and different isolates from the incubated plates were sub - cultured on fresh Nutrient agar, MacConkey agar and Blood agar plates using streak – plate method (13), to obtain pure cultures. The pure cultures isolated were sub – cultured on Nutrient agar slants and incubated at 37$^{O}$C for 24 hours. They were then stored in the refrigerator, until required for identification and susceptibility tests. All organism were identified using the method of Chesbrough, (4).

**Antibiotics susceptibility testing.**

The antibiotic discs(oxoid,India) selected for this study are commercially prepared discs of conventional antibiotics commonly used in Imo State for treatment of common infections.

## RESULTS

A total of 520 respondents (280 patients from Imo State University Teaching Hospital Orlu and 240 patients from Federal Medical Centre Owerri),288 males and 116 females were selected for this study. Of this number, 496 (95.4%) respondents completed and returned their questionnaires, 404 (77.7%) respondents accepted for wound swab to be collected from them in this study comprised of 156 traders, 108 Civil servants, 76 artisans, 48 students and 16 Applicants/Housewives( table 3). Of the 404 wound swab collected and examined, 52 (12.9%) had concomitant infections of 2 bacterial species each and 352 (87.1%) had single bacterial infection each. Generally 5 species of bacteria: *Staphylococcus aureus, Streptococcus pyogenes, Pseudomonas aeruginosa, Proteus mirabilis* and *Escherichia coli* were isolated from the patients. A total of 456 bacterial isolates were obtained. The most prevalent isolate (32.7%) is *Pseudomonas aeroginosa* and the least prevalent bacteria (12.9%) is *Streptococcus pyogenes.*

Seventy six out of the four hundred and four patients who responded to collection of wound swab were aged 18 – 25 years, 176 were aged 26 – 35 years, 116 were aged 36 – 45 years and 36 were aged 46 years and above. Amongst the age group 18 – 25 years, a total of 48 males were examined, which includes 20 (41.7%) that were infected by *Staphylococcus aureus,* 12 (25%) by *Escherichia coli,* 8 (16.7%) by *Pseudomonas aeruginosa,*8 (16.7%) by *Proteus mirabilis,* and 8 (16.7%) were infected by *Streptococcus pyogenes*. Twenty eight females were examined, 12(42.9%) were infected by *Proteus mirabilis, while* 4(14.3%), 4 (14.3%),4 (14.3%) and 12 (42.9%) were infected by *Streptococcus pyogenes, Staphylococcus aureus, Escherichia coli* and *Pseudomonas aeruginosa*. Amongst the age group 26 – 35 years, 120 males and 56 females were examined. Of this number, 28 (23.3%) males and 28 (50%) females were infected by *Staphylococcus aureus,* 28 (23.3%) males and 24 (42.9%) females were infected with *Pseudomonas aeruginosa,* 28 (23.3%) males and 4 (7.1%) female were infected by *Proteus mirabilis,* 24 (20%) males and 8 (14.3%) females were infected by *Streptococcus pyogenes* and 16 (13.3%) males and 8 (14.3%) females were infected by *Escherichia coli*(Table 1).

Table 2, shows the age of wound – related infection which revealed that out of 12 people with wounds aged 0 - 7 days; 8 (66.7%) were infected with *Proteus mirabilis* while 4 (33.3%) were infected with *Staphylococcus aureus.* A total of 120 patients had wounds aged 8 – 21 days, out of which 44 (36.7%) of the wounds were infected with *Pseudomonas aeruginosa,* 32 (26.7%) with *Staphylococcus aureus,* 20 (16.7%) *Streptococcus pyogenes,* 16 (13.3%) *Escherichia* coli and 16 (13.3%) were infected with *Proteus mirabilis*. 200 patients had wounds aged 3 – 4 weeks. Of this number, 72 (36%) were infected with *Staphylococcus aureus,*while 64 (32%) had *Pseudomonas aeruginosa,* 44 (22%), 28 (14%), and 24 (12%) were infected with *Escherichia coli, Proteus mirabilis* and *Streptococcus pyogenes.* Also 68 patients had wounds aged 1 – 3 months and 4 patients (100%) had wound aged 4 – 6 months (Table 2).

The on – patients department had 320 patients and 84 out – patients. Of this numbers, 104 (32.5%) in patients and 24 (28.5%) out – patients were infected with *Staphylococcus aureus,* 64 (20%) in patients and 16 (19%) out – patients were infected with *Escherichia coli,* 44 (13.8%) in patients and 20 (23.8%) out – patients were infected with *Proteus mirabilis,* 112 (35%) in patients and 20 (23.8%) out – patients were infected with *Pseudomonas aeruginosa* and 44 (13.8%) in patients and 8 (9.5%) out – patients were infected with *Streptococcus pyogenes* as shown on Table 4. Also among those patients examined, 280 (69.3%) had wounds located on their legs, 104 (25.7%) had wounds located on their hands while 20 (5%) had their wounds located on the chest /Neck region (Table 5). The antibiotic susceptibility of the test isolates growth inhibitory effects were as shown in Table 6.

**TABLE 1: AGE AND GENDER – RELATED PREVALENCE OF WOUND INFECTION.**

| **Age (years)** | **Sex** | **Sample** | **Numbers Infected with these organism (%)** | | | | |
|---|---|---|---|---|---|---|---|
| | | | ***S. aureus*** | ***E. coli*** | ***P.aeruginosa*** | ***P.mirabilis*** | ***S. pyogenes*** |
| **18 – 25** | Male | 48 | 20 | 12 | 8 | 8 | 8 |
| | Female | 28 | 4 | 4 | 12 | 4 | 4 |
| **26 – 35** | Male | 120 | 28 | 16 | 28 | 28 | 24 |
| | Female | 56 | 28 | 8 | 24 | 4 | 8 |
| **36 – 45** | Male | 88 | 28 | 24 | 44 | 8 | 4 |
| | Female | 28 | 8 | 8 | 4 | 4 | 4 |
| **46-above** | Male | 32 | 12 | 8 | 12 | 4 | 0 |
| | Female | 4 | 0 | 0 | 0 | 4 | 0 |
| **Total** | | **404** | **128 (31.7%)** | **80 19.8%)** | **132 (32.7%)** | **64 (15.8%)** | **52 (12.9%)** |

**TABLE 2: AGE OF WOUND – RELATED PREVALENCE OF INFECTION**

| **Age of wound** | **Number Exam.** | **Number Infected (%)** | | | | |
|---|---|---|---|---|---|---|
| | | ***S. aureus*** | ***E. coli*** | ***P.aeruginosa*** | ***S. pyogenes*** | ***P. mirabilis*** |
| **0 – 7 days** | **12** | **4** | **0** | **0** | **0** | **8** |
| **8– 21 days** | **120** | **32** | **16** | **44** | **20** | **16** |
| **3–4 weeks** | **200** | **72** | **44** | **64** | **24** | **28** |
| **1-3months** | **68** | **16** | **20** | **24** | **8** | **12** |
| **4-6months** | **4** | **4** | **0** | **0** | **0** | **0** |
| **Total** | **404** | **128(31.7%)** | **80 (19.8%)** | **132 (32.7%)** | **52(12.9%)** | **64(15.8%)** |

**TABLE 3: OCCUPATIONAL RELATED PREVALENCE OF INFECTION AMONGST ORTHOPAEDIC WOUND PATIENTS.**

| Occupation | No. Exammined | Number infected (%) | | | | |
|---|---|---|---|---|---|---|
| | | *S. aureus* | *E. coli* | *P.aeruginosa* | *S.pyogenes* | *P.mirabilis* |
| Civil Servant | 108 | 36 | 24 | 40 | 8 | 20 |
| Trader | 156 | 56 | 32 | 36 | 28 | 20 |
| Student | 48 | 12 | 8 | 8 | 8 | 12 |
| Applicant/House wife | 16 | 12 | 4 | 4 | 4 | 4 |
| Artisan | 76 | 12 | 12 | 44 | 4 | 8 |
| **Total** | **404** | **128 (31.7%)** | **80 (19.8%)** | **132 (32.7%)** | **52 (12.9%)** | **64 (15.8%)** |

**TABLE 4: PATIENTS' STATUS RELATED PREVALENCE OF INFECTION.**

| Patients' Status | Number Examined | Number Infected (%) | | | | |
|---|---|---|---|---|---|---|
| | | *S. aureus* | *E. coli* | *P.mirabilis* | *P.aeruginosa* | *S. pyogenes* |
| In-Patients | 320 | 104 | 64 | 44 | 112 | 44 |
| Out - Patients | 84 | 24 | 16 | 20 | 20 | 8 |
| **Total** | **404** | **128 (31.7%)** | **80 (19.8%)** | **64 (15.8%)** | **132 (32.7%)** | **52 (12.9%)** |

**TABLE 5: WOUND LOCATION – RELATED PREVALENCE OF INFECTION.**

| Location of Wound | Number Examined | Number Infected (%) | | | | |
|---|---|---|---|---|---|---|
| | | *Staphlococcus aureus* | *Escherichia coli* | *Pseudomonas aeruginosa* | *Streptococcus* pyogenes | *Proteus mirabilis* |
| **Legs** | **280** | **104** | **64** | **80** | **36** | **40** |
| **Hand** | **104** | **16** | **16** | **40** | **16** | **24** |
| **Chest / Neck** | **20** | **8** | **0** | **12** | **0** | **0** |
| **Total** | **404** | **128(31.7%)** | **80(19.8%)** | **132(32.7%)** | **52(12.9%)** | **64(15.8%)** |

**TABLE 6: ANTIBIOTIC SUSCEPTIBILITY PATTERN OF TEST BACTERIAL ISOLATES**

| Isolates | No Examined | Mean Zone of Growth Inhibition (mm) | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Zn | Azy | Au | Avf | Sf | Lv | Gax | Cf | S | Apx | CN | N |
| *S.aureus* | 128 | 18.6 | 18.2 | 1.4 | 16.3 | 14 | 20.1 | 8.9 | 22.6 | 10.2 | 0.5 | 6.3 | 0 |
| *S.pyogenes* | 52 | 18.6 | 20.2 | 0 | 15 | 8.2 | 16.9 | 4 | 24 | 16.4 | 0 | 3.4 | 0 |
| *P. aeruginosa* | 132 | 18.6 | 17.4 | 1.3 | 12.6 | 10.8 | 18.4 | 8.4 | 22.7 | 15.1 | 0 | 3.3 | 0 |
| *P.mirabilis* | 64 | 16.4 | 20.6 | 2.4 | 15.9 | 15.3 | 20.8 | 12.3 | 22.1 | 6.5 | 3.4 | 9.9 | 0 |
| *E. coli* | 80 | 21.3 | 18.5 | 3.1 | 15.7 | 14 | 19.1 | 7.6 | 24.1 | 7 | 3.2 | 10.6 | 0.7 |

KEY: Zn =Zinnat (cefuroxime), Azy = Azycin (Azithromycin), Au = Augmentin (Amoxycillin/Clavulanate potassium), Avf = Avicef. (Ceftriaxone), Sf = Sparflox, Lv = Levitol (Levofloxacin), Gax = Gaxin (Ofloxacin), Cf = Ciprofloxacin, S = Streptomycin, Apx = Ampiclox, Cn = Gentamycin, N = Nitrofurantoin.
*Sa = S. aureus, Sp= S.pyogenes, Pa= P.aeruginosa, Pm = P. Mirabilis,* Ec = E.coli

## DISCUSSIONS

Since wound colonization is most frequently polymicrobial, involving numerous microorganisms that are potentially pathogenic, any wound is at some risk of becoming infected. In the event of infection, a wound fails to heal, the patient suffers increased trauma, treatment costs rise, and general wound management practices become more resource demanding ( 2). In this study only five species of bacterial isolates which includes; *Staphylococcus aureus, Streptococcus pyogenes, Pseudomonas aeruginosa, Proteus mirabilis* and *Escherichia coli* were isolated from the patients, with the most prevalent isolate (32.7%) been *Pseudomonas aeroginosa* and the least prevalent bacteria (12.9%)

been *Streptococcus pyogenes*. Though in this study no fungi study was carried out, this above reports confirms the statement of Zaklad, et al,(15), when they stated that bacteria is a pathogenic factor in orthopaedic surgery. This trend of results could be as result of patients waiting for a long time before seeking for medical attention and as such leading to heavy growth of bacteria and mixed infection.
According to these authors(12) wound infection has 3 stages, early stage, which is less than 2 weeks, delayed stage, which is 2- 10 weeks and late stage, which is more than 10 weeks. The result of their bacteriological analysis showed that *Pseudomonas aeruginosa* followed by *Staphylococcus aureus* are the leading causative agents of orthopaedic wounds at National Orthopaedic Hospital Enugu (12).
In this study, it was observed that 156 patients were traders with the highest bacterial infections. Also the highest number of organisms isolated were *Staphylococcus aureus 56* (35.9%), while the least was *Proteus mirabilis* 20 (12.8%). The least infection was in applicants/housewives, 16 patients, having *Staphylococcus aureus* 12 (7.5%) and the least organism were *Proteus mirabilis* 4 (2.5%). *Pseudomonas aeruginosa* had the highest infection rate with 32.7% while *Streptococcus pyogenes* had the least infection rate of 12.9%, therefore supporting the work Mehta,et.al.,(11), when they reported that during the period from 2002 to 2005 *Pseudomonas* species was the commonest pathogen isolated (51.5%) followed by *Acinetobacter* species (14.28%), *Staphylococcus aureus* (11.15%), *Klebsiella* species (9.23%) and *Proteus* species (2.3%). When they also compared the results of the previous five years i.e., 1997 to 2002 in their study, *Pseudomonas* species was still the commonest pathogen in the burns unit (11). Also, the result in this study were in line with the works of (1), when they stated that orthopedic wounds are more in people that engaged in field works than indoor jobs. From this study, it was seen that the orthopaedic wounds on the legs had the highest infection rate of *Staphylococcus aureus* 104 (37.1%), revealing that most orthopaedic wounds occurs on the legs probably due to accidents are mostly colonized by this organisms. This might be the reason why Adebayo, et al (1) said *Staphylococcus aureus* dominated the acute soft tissue infection (26.3%) that include accidents, boils, abscesses and necrotizing infection (1). Furthermore, (3) reported the presence of pure *Streptococcus pyogenes* in two patients and a mixed predominance of *Peptostreptococcus* spp., *S. pyogenes*, *B. fragilis*, *C. perfringens*, *E. coli*, and *Prevotella* spp. in the others. Potentiation of infection by microbial synergistic partnerships between aerobes, such as *S. aureus* and *S. pyogenes*, and nonsporing anaerobes has been recognized in various types of nonclostridial cellulitis and necrotizing fasciitis (3).
Further analysis showed that males and females within the age 26-35 years old had the highest prevalence of orthopaedic wounds, while the men were mainly involved in orthopaedic wounds more than females probably because of occupational involvement.
In this study, Ciprofloxacin (ciproxin) exhibited the highest growth inhibitory effect on *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Streptococcus pyogenes*, *Proteus mirabilis* and *Escherichia coli* in decreasing order. It was observed that Nitrofurantoin did not exhibit any growth inhibitory effect on the test isolates except one strain of *Escherichia coli.* Analysis showed that *Streptococcus pyogenes were resistant to* Ampiclox and Augmentin. Ampiclox exhibited the lowest growth inhibitory effect on *Staphylococcus aureus,* Augmentin exhibited the lowest growth inhibitory effect on Proteus *mirabilis* and *Pseudomonas aeruginosa* while Nitrofurantoin exhibited the lowest on *Escherichia coli.* This reports show that the third generation cephalosporins were also very effective in the treatment of wound colonized infections such as this orthopaedic wounds studied, as Muhammed, et al, (12), in their study reported that all the infections they studied were treated with the prophylactic antibiotic of first generation cephalosporin such as cefazoline. Though, according to a report (2), antibiotic susceptibility of wound isolates observed in the laboratory cannot always be related directly to the clinical situation since the in vitro and in vivo conditions vary considerably. They further stated that the number of organisms at the infected site may be significantly different from the standard inoculum size used in vitro, the wound pH is likely to differ from test pH, and the pharmacokinetics and conditions at the infected site are also likely to influence microbial susceptibility in vivo (2).

## CONCLUSION

The results obtained from this study revealed that a variety of opportunistic and pathogenic bacteria colonized orthopedic wounds at Imo State University Teaching Hospital Orlu and Federal Medical Center Owerri both in Imo state. These infections caused an increase admission to hospitals as well as

increase in the cost of therapy, which can lead to increased morbidity and mortality in patients, economic loss and reduction of man power to the society. The high diversity of organism and poor susceptibility pattern signifies the need for proper infection control and laboratory investigation of all patients presenting with orthopedic wound infection. The majority of the bacteria isolates were resistant to most of the antibiotics used in this study. It becomes imperative that the use of antibiotics should not be abused and should be used only on doctor's prescription. Furthermore, the Government should enforce laws to reduce road accident and there should be public enlightenment on the need to seek medical help as soon as one has orthopedic wound to avoid contamination and complication due to bacterial infections.

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**REFERENCES**

1. Adebayo, O. S., Deboye, O. K., Emiola, A. R.,2003, Wound infections: In two health institution in Ile-Ife, Nigeria. Results of a cohort study. J. of Wound Management. *49, 52-57.*
2. Bowler, P.G., Duerden, B.I., Armstrong, D. G.,2001, Wound Microbiology and Associated Approaches to Wound Management. Clin. Microb. Rev. *14, 244-269.*
3. Brook, I. ,1998, Aerobic and anaerobic microbiology of infections after trauma in children. J. Accid. Emerg. Med.15:162–167.
4. Chesbrough,M.,2002, *Medical Laboratory Manual for Tropical countries.Microbiology.* Tropical Health Technology/Butterworths and Co Ltd. Cambridge/Sevanaks. 2 ,54-65.
5. Cruse, P.J. ,1992,Surgical wound infections in: wonsiewiez .Med. Journal *(ed). Infectious Diseases.* Philadelphia, W.B Saunder Co. p.758-764.
6. Edward, C., Counsell, C., Boulton, C., Moran, G .,2008, Early infection after hip fracture surgery, Risk factors, cost and outcome. J. Bone Joint Surg. 90B,770-777.
7. Elizabeth, M. A. ,2003, *Oxford Concise Dictionary.* Sixth edition, Oxford University Press .Great Britian. p. 86- 746.
8. Giles, J. T., Bartlett, S. J., Gelber, A. C., Nanda, S. ,2006, Introduction of Post Operative Infection. U.S.A. p. 345-351.
9. Idowu,O.J., Onipede, A.O., Orimolade, A.E., Akinyoola, L.A., Babalola,G.O. ,2011, Extended-spectrum Beta-lactamase Orthopedic Wound Infections in Nigeria. J. Glob. Infect. Dis. 3(3), 211–215.
10. Jawetz, E., Melnick, J. I., Adelberg, E. A. ,2007, Medical Microbiology. 24th edition. The Mcgraw- Hill companies, Inc., United States of America, California p. 550-554.
11. Mehta, M., Dutta, P., Gupta, V., 2007 , Bacterial isolates from burn wound infections and their antibiograms: A eight-year study. India J. of Plastic Surg. 40(1), 25-28.
12. Muhammad, S. K., Saifur, R., Mian, A., Babar, S., Shahid, S. ,2008, Infect. in Othorpaedic Implant Surgery, *Its Risk Factors and Outcome. 20 (1), 23-27.*
13. Obiajuru, I. O, C., Ozumba, U. C.,2009, Laboratory Methods for Medical Microbiology & Parasitology. Lifeway Amalgamations. Owerri. p.183.
14. Reichardt, P. , 2009, The clinical case for using hyperbaric oxygen therapy in the treatment of diabetic wound. Journal Featuring Articles About Wound Care And Related Research, PMCA *99, 214-225.*
15. Zaklad, D., Leczema, C., Medyczny, W., 2006, Fungi and Bacteria as a Factor in Wound Healing in Patients After Orthopaedic surgeries. p.78-89.