

## Bacterial and Antibiotic Sensitivity Pattern in Secondary Peritonitis

### Abstract

**Background:** Peritonitis is inflammation of the peritoneum usually as a result of a localized or generalized infection. Secondary peritonitis which is the most common type follows an infective process in a visceral organ. The role of peritoneal cultures and use of antibiotics effective against culture results remain controversial. **Objectives:** This study was conducted to determine the bacterial and antibiotic sensitivity pattern in patients with secondary peritonitis. It also compared the use of empirical antibiotics and culture-sensitive antibiotics with outcomes of patients with secondary peritonitis. **Materials and Methods:** A prospective randomized clinical study was conducted. Five millilitres of peritoneal fluid was sampled intra-operatively, and microscopy, culture, and sensitivity testing was performed in patients with secondary peritonitis. The patients, randomized into two groups, had antibiotics administered for 7 days. The first group had empirical antibiotics throughout (Ceftriaxone + Metronidazole), whereas the second group had empirical antibiotics (Ceftriaxone + Metronidazole) for the first 2 days and antibiotics according to the sensitivity report for the remaining 5 days. The *post-hoc* analysis was also done on a third group, who, even though were randomized to either groups, had no growth on culture of peritoneal fluid. **Results:** The commonest pathogens identified from the peritoneal culture of the participants were *Escherichia coli*, *Klebsiella pneumoniae*, *Anaerococcus* group, and *Bacteroides fragilis*. Complications including mortality were significantly higher in those who received empirical antibiotics than those who received culture-sensitive antibiotics. **Conclusion:** The outcome of antibiotics administration in patients with secondary peritonitis with a positive culture was better in those who received culture-sensitive antibiotics than those who received empirical antibiotics.

**Keywords:** Antibiotic sensitivity pattern, bacterial culture, outcomes, secondary peritonitis

### Introduction

Peritonitis is inflammation of the peritoneum which lines the abdominal cavity and its contained organs. The peritoneum reacts to various pathological stimuli which could be infectious, chemical or mechanical.<sup>[1]</sup>

Primary peritonitis results from bacterial, fungal, chlamydial, or mycobacterial infection in the absence of perforation of the gastrointestinal tract. It is usually seen in young girls between 3 and 10 years and occasionally in boys and results from bacteria entering the abdomen through the vagina or haematogenously from a respiratory infection. It can also be seen in patients with cirrhosis of the liver with ascites or in patients with nephrotic syndrome. Secondary peritonitis is inflammation of the peritoneum, which follows an infective process in abdominal viscera. Tertiary peritonitis, a less well-

defined entity, is characterized by persistent or recurrent infection following operative attempt to treat secondary peritonitis.<sup>[1]</sup>

Clinically, peritonitis is often classified either as local or as diffuse. Local peritonitis refers to loculi of infection usually walled off or contained by adjacent organs. Diffuse peritonitis is synonymous with generalized peritonitis and involves the entire abdominal cavity.<sup>[1]</sup>

The organisms that cause peritonitis are usually mixed but consist mainly of *Escherichia coli* and *Bacteroides* spp.<sup>[1]</sup> Since the demonstration of the microbial basis for peritonitis in 1887, surgeons have been looking for the chemotherapeutic means to treat this disease.<sup>[2]</sup> Although the primary therapeutic modality for secondary peritonitis is source control via surgery, antimicrobial agents play an important role.<sup>[3]</sup>

The role of peritoneal cultures and use of antibiotics effective against culture results

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**Adedoyin Babatunde Ojo,  
David Omoareghan Irabor<sup>1</sup>**

Department of Surgery,  
University College Hospital,  
Ibadan, <sup>1</sup>Department of  
Surgery, College of Medicine,  
University of Ibadan, Ibadan,  
Nigeria

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**Address for correspondence:**  
Dr. Adedoyin Babatunde Ojo,  
Department of Surgery,  
University College Hospital,  
Ibadan, Nigeria.  
E-mail: ojo.adedoyin@yahoo.com

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remain controversial. Some earlier studies revealed that culture result data rarely contributed in a useful way to the patient's post-operative outcome.<sup>[2,4]</sup> More recent studies show an improvement in patients' outcomes with antibiotics use according to culture and sensitivity.<sup>[5-7]</sup>

Empirical antibiotic therapy, which is defined as that regimen initiated at the time of operative intervention and before the availability of any culture data, should be effective against likely organisms that may be isolated.<sup>[2]</sup> A combination of a third-generation cephalosporin and metronidazole has been recommended for secondary peritonitis. Other recommended combinations include penicillin/beta-lactamase inhibitor and ciprofloxacin/clindamycin.<sup>[4]</sup>

This study aimed to culture peritoneal fluid in patients with secondary peritonitis and also compared the outcomes of treatment with empirical antibiotics and culture-sensitive antibiotics.

## Materials and Methods

This was a prospective randomized clinical study. It was a parallel trial design with a 1:1 allocation ratio. A total of 60 consecutive patients aged 16 and above with secondary peritonitis were recruited, with 30 subjects in each arm.

A simple randomization technique was used in the assignment of participants, and this was achieved with a randomization schedule generated with an online computer software.

Patients in Group A received empirical antibiotics (Ceftriaxone + Metronidazole) throughout for 7 days, whereas patients in Group B received empirical antibiotics (Ceftriaxone + Metronidazole) for the first 2 days and antibiotics according to peritoneal culture sensitivity data for the remaining 5 days. The whole process of generation and implementation of randomization was done by the principal investigator of this study, and he was not blinded to the modality of antibiotics used in the post-operative management of the patients. The assessor of incision healing (intern) was blinded to which participant had empirical or culture-sensitive antibiotics. The *post-hoc* analysis was done on a third group (Group X), who, even though was randomized to either group, had no growth on culture of peritoneal fluid and had significant differences in characteristics and outcomes.

The patients were operated upon following adequate resuscitation. Five millilitres of peritoneal fluid was collected aseptically immediately after opening the peritoneum. It was examined macroscopically for colour, odour, and quality (clear, cloudy/purulent, or faecal). It was collected in sterile universal bottles and transported to the microbiology department for microscopy, culture, and sensitivity. Five millilitres of peritoneal fluid was collected in Robertson's cooked meat medium for back-up culture.

Blood agar, MacConkey agar, and Chocolate agar were used for aerobic culture, whereas Bacteroides Bile Esculin agar and Neomycin-Vancomycin laked Blood agar were used for anaerobic culture. Sensitivity testing was carried out on the aerobic isolates using standardized disc diffusion technique with Mueller–Hinton agar. The diameter of zone of inhibition was measured and interpreted using the Clinical and Laboratory Standard Institute guideline (2018).<sup>[8]</sup> Media inoculation was done on the first day. If growth was present on the second day, the organisms were identified and an antibiotic sensitivity was done. Results of organisms isolated and its antibiotic sensitivity pattern were ready for all patients on the third day. All these were done by one laboratory scientist in order to minimize observer bias.

All patients had the appropriate surgical intervention. Thorough cleansing of the peritoneal cavity with volumes of warm saline was done, and the laparotomy wound was closed.

The wound was assessed routinely on the seventh post-operative day by an intern using the Southampton scoring system.<sup>[9]</sup> The intern was blinded as to which group (and antibiotic modality) the patient belonged. Wound complications required more frequent assessment, and appropriate treatment was provided to the patients.

Ethical approval was obtained, and utmost confidentiality was observed throughout the conduct of this research.

## Results

Out of a total of 60 participants in this study, 30 patients were allocated randomly to each group of the study. The *post-hoc* analysis was done on a third group (Group X), who, even though were randomized to either group, had no growth on culture of peritoneal fluid. Hence, the study consisted of Group A who had positive culture and received empirical antibiotics with a number of 23 participants, Group B who had positive culture and received culture-sensitive antibiotics when sensitivity became available after 2 days of empirical antibiotics with a number of 21 participants, and Group X who had no growth and received empirical antibiotics with a number of 16 participants.

Of the 60 patients operated upon during the study period, 44 (73.3%) were males whereas 16 (26.7%) were females giving a sex ratio of 2.8:1. The ages of the patients ranged from 16 to 66 years with a median age of 35.5 and a modal class of 21–30.

The commonest symptoms were abdominal pain (78.3% of the patients), abdominal distension (50%), vomiting (48.3%), and fever (31.7%). Other symptoms were anorexia (31.7%) and constipation (21.7%). The mean duration (SD) of the earliest symptom in each group was  $5.1 \pm 2.0$  days for Group A (empirical),  $5.6 \pm 1.6$  days for Group B (culture-sensitive), and  $2.9 \pm 1.5$  days for Group X (no growth).

( $P = 0.002$ ). The mean duration of symptoms for all patients was  $4.7 \pm 4.2$  days.

Fifty-six (93.3%) patients had no co-morbidity when using the Elixhauser co-morbidity index. Two (3.3%) had uncomplicated hypertension, one (1.7%) had uncomplicated diabetes, and one (1.7%) had peptic ulcer disease.

The patients were of comparable American Society of Anaesthesiology (ASA) class. Majority (56.7%) of all patients were of ASA III, 25% were ASA II, 10% were ASA I, and the remaining 8.3% were ASA IV. The average time taken from when a diagnosis of secondary peritonitis was made and the decision taken to operate on the patient to the time when an incision was made was  $14.3 \pm 9.9$  h.

A large portion (56.7%) of the patients had purulent exudate, 28.3% had haemorrhagic exudate, and 8.8% had clear exudate. Five per cent had faecal exudate, and 1.7% had bilious exudate.

The commonest diagnoses in the descending order were perforated peptic ulcer disease (23.3%), penetrating abdominal injury (21.7%), complicated appendicitis—perforated and gangrenous (20%), perforated typhoid ileitis (8.3%), blunt abdominal injury (6.7%), strangulated adhesive intestinal obstruction (5%), and iatrogenic bowel injuries (3.3%). Similarly, the commonest procedures done were closure of gastric and duodenal perforations (23.3%), appendectomy (20%), and closure of ileal perforation (15%). Ileal resection and anastomosis, right hemicolectomy, repair of hepatic laceration, and Hartmann's colostomy round off the procedures done.

Of the 60 participants, 44 had a positive culture. The common isolates are shown in Table 1. A single pathogen was isolated in 28 (63.6%) participants, whereas 16 (36.3%) cultures identified multiple pathogens.

**Table 1: Pathogens isolated from peritoneal culture of participants**

Common isolates	n (%)
<b>Aerobes</b>	
<i>Escherichia coli</i>	22 (36.7)
<i>Klebsiella pneumoniae</i>	19 (31.7)
<i>Candida albicans</i> *	11 (18.3)
<i>Staphylococcus aureus</i>	9 (15)
<i>Enterococcus faecalis</i>	8 (13.3)
<i>Enterobacter cloacae</i>	7 (11.7)
<i>Streptococcus</i> spp.	7 (11.7)
<i>S. albus</i>	6 (10)
MRSA	6 (10)
<b>Anaerobes</b>	
<i>Anaerococcus</i> group	10 (16.7)
<i>Bacteroides fragilis</i>	9 (15)
<i>Peptococcus</i> spp.	1 (1.7)

\*Fungus

The sensitivity rate for the commonly used antibiotic combinations was determined in relation to the isolated pathogens and arranged in the descending order [Table 2]. Vancomycin or meropenem combinations with metronidazole showed the strongest efficacies. Amoxicillin or ciprofloxacin or gentamicin combinations with metronidazole, though strong, were inferior.

Of the 21 patients who received antibiotics according to the sensitivity profile, eight received Ciprofloxacin + Metronidazole, six received Gentamicin + Metronidazole, and three had Meropenem + Metronidazole. Two patients received Vancomycin alone and two received Piperacillin/Tazobactam alone. No patient randomized to this group had a result showing sensitivity to Ceftriaxone + Metronidazole. All surviving participants received antibiotics for 7 days. No patient in this study gave a clinical history of allergy to any antibiotics, and no adverse reaction due to drug toxicity was noted in any patient during the course of their hospital stay.

Return of bowel function was quicker in individuals with no growth on culture of peritoneal fluid (3.1 days) when compared with those with a positive culture (4.3 days), and this was statistically significant ( $P = 0.043$ ). It was defined by when the participant passed flatus or faeces, and it signalled the commencement of graded oral feeds.

Repeated-measures analysis of variance curve showing pulse rate revealed a significant reduction over the course of hospital stay with rates close to normal on the seventh post-operative day. This reduction was most marked in individuals with a negative culture and better in individuals who had culture-sensitive antibiotics compared with those who had empirical antibiotics.

A review of the haematological indices showed that 15% of all patients had anaemia pre-operatively and 56.7% had leucocytosis. All the patients showed a log decline in white blood cell count when the blood count was repeated prior to discharge, with the decline sharpest in those who received culture-sensitive antibiotics.

The incidence of complications, as demonstrated in Figure 1, was higher in the group that had a positive culture and received empirical antibiotics. A larger

**Table 2: Efficacy of selected antibiotics combination against all pathogens isolated**

Antibiotics combination	Efficacy (%)
Vancomycin + Metronidazole	94
Meropenem + Metronidazole	93
Levofloxacin + Metronidazole	88
Piperacillin/Tazobactam + Metronidazole	87
Amoxicillin/Clavulanic acid + Metronidazole	79
Ceftriaxone + Metronidazole	76
Gentamicin + Metronidazole	73
Ciprofloxacin + Metronidazole	71
Amoxicillin + Metronidazole	70

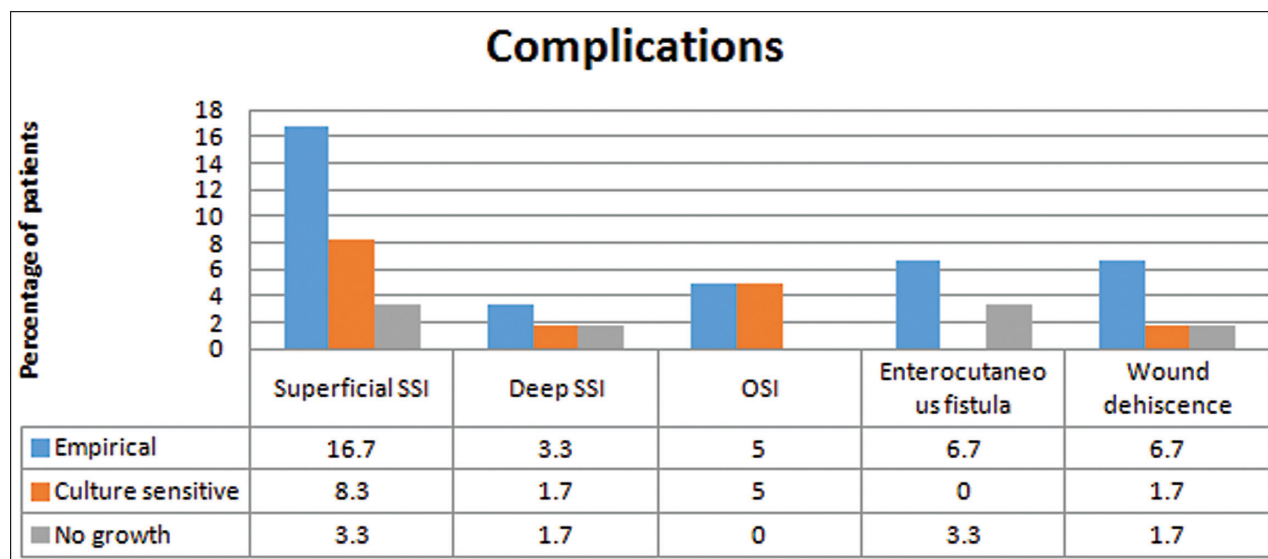


Figure 1: Complications of surgical intervention (SSI = Surgical Site Infection, OSI = Organ/Space Infection)

percentage of them (10%) required admission into the intensive care unit compared with only 1.7% in the other groups ( $P = 0.312$ ). Similarly, the use of ventilator and inotropic support was higher in this group (Group A) ( $P = 0.514$ ).

The need for a relaparotomy (5%) and secondary wound closure (3.3%) was only in Group A ( $P = 0.231$ ). The mean hospital stay (SD) was 15.8 (13.7) days for Group A, 9.2 (5.1) days for Group B, and 8.5 (4.7) days for Group X. The median hospital stay for all patients was 8 days.

Twenty-seven patients (45%) had surgical site infection (SSI) with the most common complication being superficial incisional SSI. The pathogens isolated in peritoneal fluid of these patients with complications of SSIs were *E. coli* (14 patients), *K. pneumonia* (11 patients), and *B. fragilis* (2 patients).

The incidence of complications was higher in those who received empirical antibiotics; however, this difference was not statistically significant ( $P = 0.253$ ).

There was a significant difference between the hospital stay of the patients in the three groups ( $P = 0.036$ ). Patients who received empirical antibiotics had a mean hospital stay duration of 18.2 days, whereas those who received culture-sensitive antibiotics or had no growth had a mean hospital stay of 9.2 and 8.5 days, respectively.

Seven patients required immediate post-operative intensive unit care. Four had perforated typhoid ileitis, whereas three had perforated peptic ulcer disease.

Three participants, all in Group A, died giving a mortality rate of 5%. Two had perforated typhoid ileitis and one had multiple small bowel perforations following a gunshot injury. The pathogens isolated from culture of their peritoneal fluid were *K. pneumoniae*, *Streptococcus* spp.,

Table 3: Comparison of survivors and non-survivors

	Survivors (n = 57)	Non-survivors (n = 3)
Mean ASA (SD)	2.6 (0.8)	3.3 (0.6)
Inotropic support, n (%)	1 (1.8)	1 (33.3)
Ventilatory support, n (%)	1 (1.8)	1 (33.3)
ICU stay, n (%)	5 (8.8)	2 (66.7)
Positive culture, n (%)	41 (71.9)	3 (100)
Post-operative enterocutaneous fistula, n (%)	4 (7.0)	2 (66.7)

and *E. cloacae*. Using the multivariate analysis, mortality was closely associated with high ASA class, use of inotropic and ventilator support post-operatively, intensive care unit stay, a positive culture, and post-operative enterocutaneous fistula. Comparison of survivors and non-survivors is highlighted in Table 3.

### Discussion

The main finding of this study was that the commonest pathogens identified aerobically from peritoneal culture of participants were *E. coli* and *K. pneumoniae*. Anaerobic culture showed *Anaerococcus* group and *B. fragilis* as the most predominant pathogens. Secondary peritonitis has been described as a polymicrobial infection.<sup>[4,10]</sup> However, a single pathogen was isolated in a larger percentage of the participants of this study.

The clinical symptoms of secondary peritonitis include abdominal pain, abdominal distension, vomiting, fever, anorexia, and constipation. The mean time lapse between onset of symptoms and presentation to the hospital for all patients was  $4.7 \pm 4.2$  days, and this was comparable to  $5.4 \pm 3.7$ <sup>[11]</sup> and  $5.5 \pm 3.5$  days<sup>[12]</sup> in other studies. Patients with peritonitis who had no growth on peritoneal culture presented earlier and this was statistically significant.



The antibiotics combination regimens with the strongest efficacy against all pathogens isolated in this study are Vancomycin + Metronidazole and Meropenem + Metronidazole. Levofloxacin + Metronidazole, Piperacillin/Tazobactam + Metronidazole, Amoxicillin/Clavulanic acid + Metronidazole, and Ceftriaxone + Metronidazole also show strong efficacies. The best antimicrobial coverage in other studies was Vancomycin and/or Meropenem.<sup>[13,14]</sup> Combination antimicrobial therapy involving Meropenem and Vancomycin can be administered for patients in septic shock with onset of organ failure.<sup>[13]</sup> However, antimicrobial therapy should be based on local epidemiology, clinical severity of infection, and infection source.<sup>[5,13]</sup>

In this study, post-operative infectious complications were associated with isolation of *E. coli*. *E. coli* and *B. fragilis* have been described as being the main pathogens for these complications.<sup>[6]</sup>

The prevalence of invasive fungi has been described in secondary peritonitis. *C. albicans* was detected in 18% of the study participants. No association has been described between their presence and mortality and hence coverage is not recommended.<sup>[13]</sup>

Ileus is a significant factor influencing the length of hospital stay after abdominal surgery and has great implications for patients and resource utilization.<sup>[15]</sup> The mechanisms responsible for ileus include the inflammation of the peritoneum by microbial toxins or chemical irritation and associated fibrinous adhesions. Bacterial lipopolysaccharide causes ileus by initiating an inflammatory response within the intestinal smooth muscle layers and a subsequent reduction in both *in-vivo* and *in-vitro* smooth muscle contractility.<sup>[16]</sup> Return of bowel function was quicker in individuals with no growth on culture of peritoneal fluid when compared with those with a positive culture ( $P = 0.043$ ). Early return of bowel function was strongly associated with earlier discharge (on or before 7 days) in the course of this study, which agrees with the findings from studies that have evaluated the economic burden of post-operative ileus.<sup>[17,18]</sup>

Ventilatory and inotropic support and intensive care unit admission were more frequently employed in those who received empirical antibiotics compared with those who received culture-sensitive antibiotics. These indices which were predictive of adverse outcomes had been described to be worse in individuals with severe abdominal sepsis.<sup>[19]</sup>

SSI is a significant problem in the post-operative period. Of all abdominal surgeries, surgeries for peritonitis have a high frequency of SSI,<sup>[20]</sup> with rates as high as 64.2% described.<sup>[21]</sup> Twenty-seven patients (45%) had SSI with the most common complication being superficial incisional SSI.

Complications such as superficial incisional SSI, deep incisional SSI, organ/space infection, enterocutaneous

fistula, and wound dehiscence were significantly higher in those who received empirical antibiotics ( $P = 0.001$ ). The total complications were comparable in those who received culture-sensitive antibiotics and those with a negative culture. A longer hospital stay and a higher mortality rate were observed in the group that received empirical antibiotics. These findings agreed with the results obtained from studies that have compared use of empirical and culture-sensitive antibiotics in secondary peritonitis.<sup>[2,7,22]</sup>

The Southampton class was higher in participants who received empirical antibiotics. The assessment of the wound class using the Southampton scoring system was done by an intern who was blinded to the patients' groups. About 43.3% of the patients had normal wound healing (Class O) regardless of the group similar to figures obtained by Kache *et al.*<sup>[23]</sup> All participants had primary closure of laparotomy wounds.

The mortality rate in this study was 5%, and it was higher in those who received empirical antibiotics. This difference was not statistically significant and should be interpreted with caution because two participants who were randomized to the empirical antibiotics group died on the 3rd and 4th post-operative day, respectively. Since all participants received at least 2 days of empirical antibiotics, a day or two of culture-sensitive antibiotics may or may not have made a significant difference in outcomes of these two participants if they were randomized to the other group.

Independent risk factors for mortality identified in this study include ASA ( $> 3$ ), use of inotropic and ventilatory support post-operatively, intensive care unit stay, a positive culture, and post-operative enterocutaneous fistula.

## Conclusion

The commonest pathogens identified from peritoneal culture of participants are *E. coli*, *K. pneumonia*, *Anaerococcus* group, and *B. fragilis*.

The antibiotics combination regimens with the strongest efficacy against all pathogens isolated in this study are Vancomycin + Metronidazole and Meropenem + Metronidazole.

Complications such as SSI and wound dehiscence were significantly higher in those who received empirical antibiotics. Overall, prognostic indices were best in those with no growth on peritoneal culture.

## CONSORT reporting guidelines

This study adheres strictly to CONSORT guidelines, and a completed CONSORT checklist is attached to the manuscript.

## Availability of data and materials

The datasets used during this study are available on request from the corresponding author.

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Nil.

## Conflicts of interest

There are no conflicts of interest.

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