10.5005/jp-journals-10068-0005

ORIGINAL ARTICLE

# Bacteriological Profile and Antimicrobial Susceptibility Pattern of Intra-abdominal Infections: A Study from a Tertiary Care Hospital of North India

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## ABSTRACT

**Background and objectives:** Intra-abdominal infections (IAIs) are associated with significant morbidity and mortality. Pathogenic isolates and emerging resistance to commonly used antimicrobials have been a matter of concern in IAIs. In the present study, bacteriological profile and antimicrobial susceptibility of isolates from IAIs were investigated.

**Materials and methods:** A total of 145 samples (ascitic fluids, n = 56; bile, n = 20; and pus, n = 36) were collected from suspected IAI of patients reporting to the hospital and cultured. Identification of the isolates was done using standard identification protocol. Antimicrobial susceptibility was performed by Kirby-Bauer disk diffusion method and interpretation was done according to the Clinical and Laboratory Standards Institute (CLSI) guidelines.

**Results:** Of 145 samples, 112 were culture positive and 33 were sterile. Gram-negative organisms (n = 85) outnumbered the Gram-positive organisms (n = 27). Among the Gram-negative organisms, *Escherichia coli* (n = 31) was the most commonly isolated organism followed by *Klebsiella* sp. (n = 19), *Acinetobacter* sp. (n = 14), *Pseudomonas* sp. (n = 10), *Proteus* sp. (n = 5), *Citrobacter* sp. (n = 3), and *Enterobacter* sp. (n = 3). Among the Gram-positive bacteria, the most common organism was *Staphylococcus aureus* (n = 19) followed by *Enterococcus faecalis* (n = 8). Gram-negative bacilli showed significant resistance to almost all of the commonly used antibiotics. The rate of methicillin-resistant *S. aureus* (MRSA) was 36.84%.

**Conclusion:** Prompt starting of empirical antimicrobials based on the local susceptibility pattern, followed by modification of treatment in accordance with the antimicrobial susceptibility report can significantly reduce the morbidity and the mortality associated with IAIs.


**Keywords:** Emerging resistance, Empirical antimicrobials, Intra-abdominal infections.

**How to cite this article:** Garg R, Singh VA. Bacteriological Profile and Antimicrobial Susceptibility Pattern of Intra-abdominal Infections: A Study from a Tertiary Care Hospital of North India. J Gastrointest Infect 2017;7(1):21-25.

**Source of support:** Nil

**Conflict of interest:** None

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## INTRODUCTION

Intra-abdominal infections involve extensive variety of pathological conditions ranging from uncomplicated appendicitis to fecal peritonitis.[1] Intra-abdominal infections are classified into uncomplicated IAIs and complicated IAIs.[2,3] They are further classified into community-acquired IAIs and hospital-acquired IAIs. Uncomplicated IAIs involve a single organ and can be easily managed by surgical resection and antibiotics, while complicated IAIs involve more than one organ, including peritoneum, and leads to either local or diffuse peritonitis. Complicated IAIs remain an important cause of patient morbidity and are frequently associated with poor clinical prognosis. Hospital-acquired IAIs are associated with higher mortality rates because of compromised patient's immunity due to underlying illness and infections with multidrug-resistant organisms.[4] Antimicrobial treatment plays a vital role for management of critically ill patients with IAIs.[2] Initially, empirical therapy should be started based on the most frequently isolated organisms and according to the local pattern of antibiotic resistance, which should be modified to specific antimicrobials after receiving the microbiology report of antimicrobial susceptibility of the isolated organism.[4] Hence, accurate and timely identification of pathogenic organisms along with their susceptibility testing requires the attention of microbiologists along with the clinicians' diagnosis.

The threat of antimicrobial resistance to commonly used antibiotics is the major challenge faced by the clinicians nowadays in treating IAIs. As a result, growing emergence of multidrug resistance, limited availability of newer antibiotics, and scarcity of local data on antimicrobial resistance pattern of IAIs create a void for the management of IAIs. By keeping in mind the above lacunae, the present study was planned to determine the bacteriological profile and antimicrobial susceptibility pattern of isolates from IAIs.

## MATERIALS AND METHODS

### Patients and Samples

The present cross-sectional study was conducted to determine the bacteriological profile and antimicrobial

susceptibility pattern of isolates from samples obtained from IAIs in the Department of Microbiology, Maharishi Markandeshwar Institute of Medical Sciences & Research, Ambala, Haryana, India. Ethical clearance for the study was taken from the Institutional Ethical Committee. The inclusion criterion was samples from clinically suspected cases of IAIs of all ages and the exclusion criterion was noninclusion of samples from patients having infections other than IAIs.

### Processing of Samples

A total of 145 samples, viz., ascitic fluid, bile, and pus, were obtained from patients suffering from suspected IAIs like peritonitis, cholelithiasis, appendicitis, pancreatitis, and liver abscess. All the samples were cultured on blood agar and MacConkey agar and incubated at 37°C for 18 to 24 hours. Gram staining was performed on each sample. Identification of the isolates was done using standard identification protocol and other relevant biochemical tests as appropriate for the isolates.[5-7]

### Antibiotic Susceptibility Tests

Antibiotic susceptibility of the isolates was performed by Kirby-Bauer disk diffusion method and interpreted according to the CLSI guidelines, except for tigecycline for which European Committee on Antimicrobial Susceptibility Testing guidance document was followed.[8-10] Antibiotic susceptibility testing for Gram-negative organisms was put up using gentamicin, amikacin, ciprofloxacin, levofloxacin, cotrimoxazole, ceftriaxone, cefotaxime, imipenem, amoxicillin-clavulanic acid, piperacillin-tazobactam, and colistin.

Antimicrobial susceptibility testing for Gram-positive cocci was put up as follows:

- *Staphylococcus aureus*: Methicillin resistance for *S. aureus* was detected by cefoxitin disk diffusion method as recommended by CLSI guidelines,[9] and *S. aureus* showing resistance to cefoxitin was considered as MRSA. This MRSA was also tested for antimicrobial susceptibility toward cotrimoxazole, erythromycin, clindamycin, ciprofloxacin, amikacin, vancomycin, linezolid, dalfopristin/quinupristin, and tigecycline. Methicillin-sensitive *S. aureus* (MSSA) was tested for antibiotic susceptibility toward ampicillin, amoxicillin-clavulanic acid, cotrimoxazole, erythromycin, clindamycin, ciprofloxacin, amikacin, vancomycin, and linezolid.
- *Enterococcus faecalis*: Antibiotic susceptibility of *E. faecalis* was done using penicillin, ampicillin, erythromycin, tetracycline, chloramphenicol, vancomycin, linezolid as well as high-level gentamicin (120 µg) and high-level streptomycin (300 µg). Vancomycin-resistant *Enterococci* were detected based on disk diffusion results using vancomycin disk (30 µg) as per CLSI guidelines.[9] Vancomycin-resistant *E. faecalis* was subjected to linezolid and tigecycline.

## RESULTS

Of 145 samples collected from suspected IAIs, 112 showed positive culture, whereas the remaining 33 were sterile, showing a positivity rate of 77.24%. Of 112 culture-positive samples, 56 were ascitic fluids, 20 bile samples, and 36 pus samples (Table 1). Both Gram-positive and Gram-negative organisms were isolated from the processed samples. Gram-negative organisms (n = 85) outnumbered the Gram-positive organisms (n = 27). Among the Gram-negative organisms, *E. coli* (n = 31) was the most commonly isolated organism followed by *Klebsiella* sp. (n = 19), *Acinetobacter* sp. (n = 14), *Pseudomonas* sp. (n = 10), *Proteus* sp. (n = 5), *Citrobacter* sp. (n = 3), and *Enterobacter* sp. (n = 3). Among the Gram-positive bacteria, *S. aureus* (n = 19) was the most common isolate, followed by *E. faecalis* (n = 8; Table 2).

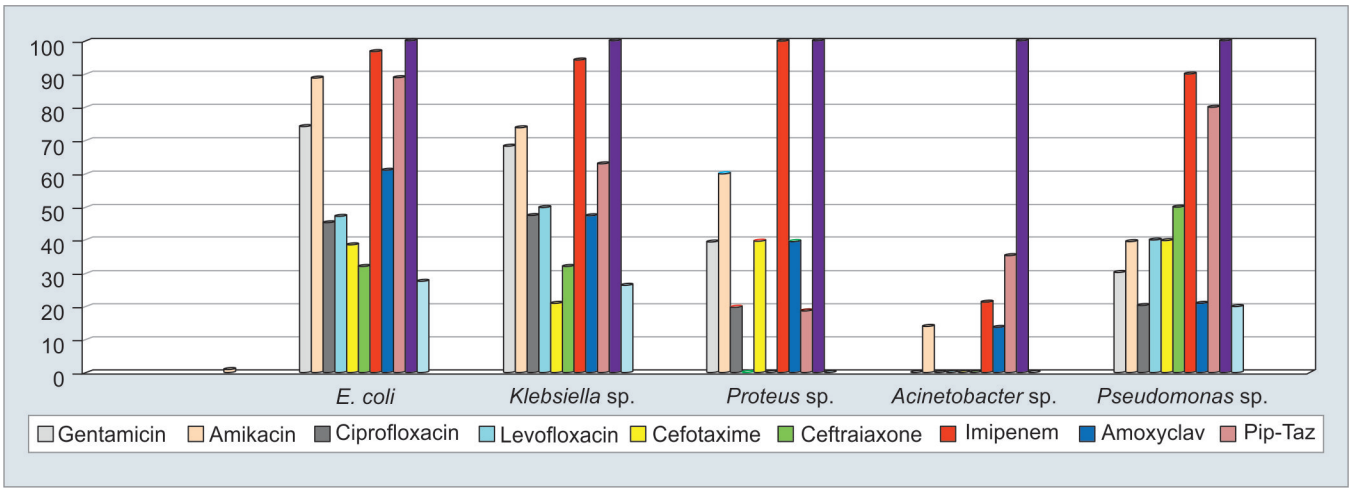
All the Gram-negative bacilli showed substantial resistance to almost all of the commonly used antibiotics. The most commonly isolated bacilli, viz., *E. coli*, showed the least sensitivity to cotrimoxazole (29.03%) followed by ceftriaxone (32.25%), cefotaxime (38.70%), ciprofloxacin (45.16%), amoxicillin-clavulanic acid (61.29%), gentamicin (74.19%), amikacin (89.09%), piperacillin-tazobactam (89.09%), and imipenem (96.77%). *Klebsiella* sp. also showed the same pattern of sensitivity. *Citrobacter* sp. and *Enterobacter* sp. showed 100% sensitivity to all the antimicrobials tested. Among nonlactose fermenters, *Acinetobacter* sp. was found to be the most resistant organism. All the 14 isolates of *Acinetobacter* sp. showed multidrug resistance and only 21.42% isolates showed sensitivity to imipenem. On the contrary, *Pseudomonas* sp. showed better sensitivity as compared with *Acinetobacter* sp. Graph 1 shows antimicrobial sensitivity pattern of Gram-negative bacilli.

**Table 1:** Samples collected from various GI infections

| *Gastrointestinal infections* | *No. of cases* | *Samples received* | *No. of samples* |
|---|---|---|---|
| Peritonitis | 60 | Ascitic fluid | 53 |
| | | Pus | 7 |
| Cholelithiasis | 19 | Bile | 19 |
| Appendicitis | 13 | Pus | 13 |
| Pancreatitis | 9 | Ascitic fluid | 3 |
| | | Pus | 5 |
| | | Bile | 1 |
| Liver abscess | 11 | Pus | 11 |
| *Total* | *112* | | *112* |

**Table 2:** Organisms isolated from various GI infections

| *Gastrointestinal infection* | *Organisms isolated* | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | *E. coli* | *Klebsiella* sp. | *Proteus* sp. | *Citrobacter* sp. | *Enterobacter* sp. | *Pseudomonas* sp. | *Acinetobacter* sp. | *S. aureus* | *Enterococcus* sp. |
| Peritonitis | 15 | 11 | 2 | 3 | 3 | 4 | 7 | 9 | 6 |
| Cholelithiasis | 6 | 2 | 1 | 0 | 0 | 3 | 3 | 4 | 0 |
| Appendicitis | 3 | 3 | 0 | 0 | 0 | 1 | 2 | 3 | 1 |
| Pancreatitis | 4 | 1 | 1 | 0 | 0 | 1 | 0 | 2 | 0 |
| Liver abscess | 3 | 2 | 1 | 0 | 0 | 1 | 2 | 1 | 1 |
| *Total* | *31* | *19* | *5* | *3* | *3* | *10* | *14* | *19* | *8* |



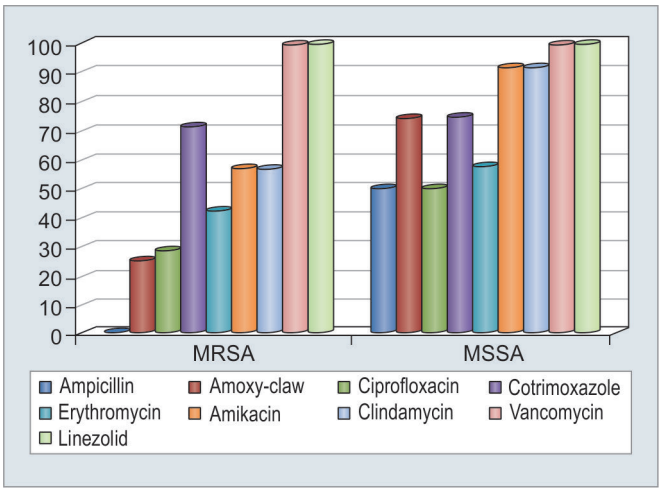

**Graph 1:** Percentage of antimicrobial susceptibility of Gram-negative bacilli isolated from IAIs

Among the Gram-positive bacteria, *S. aureus* was the most common organism isolated. The rate of methicillin resistance was 36.84% (7/19) in *S. aureus.* All the MRSA showed 100% sensitivity to vancomycin, linezolid, dalfopristin/quinupristin, and tigecycline and 71.4% to cotrimoxazole, 57.14% each to amikacin and clindamycin, 28.57% to ciprofloxacin, and 42.8% to erythromycin.

All (100%) MSSA were sensitive to vancomycin and linezolid and 91.66% each to amikacin and clindamycin. Susceptibility was 75% toward cotrimoxazole, 58.33% for erythromycin, and 50% for ciprofloxacin. The susceptibility pattern of both MSSA and MRSA is shown in Graph 2.

All the eight isolates of *E. faecalis* showed susceptibility to linezolid, while three of eight isolates showed resistance to vancomycin. Sensitivity to penicillin, ampicillin, erythromycin and tetracycline was very low, whereas 50% *E. faecalis* isolates showed susceptibility to chloramphenicol.

A high-level resistance to aminoglycosides, i.e., 25% to gentamicin (120 µg) and 37.5% to streptomycin (300 µg), was seen in three of the eight isolates. Two of the three showed combined resistance to both the high-level aminoglycosides. Three isolates showed resistance to vancomycin, but none of the isolates showed resistance to linezolid.




**Graph 2:** Antibiotic susceptibility patterns of *Staphylococcus aureus* isolated from IAIs

Vancomycin-resistant *Enterococci* were tested against linezolid and tigecycline and showed 100% susceptibility to both linezolid and tigecycline.

## DISCUSSION

The emergence of resistance to routinely used antibiotics and even to newer antibiotics has made the treatment of IAIs a real challenge for clinicians. Accurate laboratory identification of the isolates along with routine sensitivity

testing is needed. Proper collection and transport of specimens is a great challenge. Furthermore, data on local pattern of susceptibility of pathogens can help in guiding the treatment of these pathogens. Hence, clinicians and microbiologists working in tandem can go a long way in decreasing the mortality due to IAIs.

The rate of isolation of pathogenic organisms from our study was 112/145 (77.24%). Both Gram-negative bacilli and Gram-positive cocci were isolated. Among the Gram-negative bacilli, the major pathogens involved belonged to the family Enterobacteriaceae. These observations were also made by Sartelli et al.[1] Among the family Enterobacteriaceae, *E. coli* was the most common organism isolated. Studies done by Kurup et al[11] and Hawser et al[12] reported similar results. Apart from *E. coli*, organisms isolated were *Klebsiella* sp., *Proteus* sp., *Citrobacter* sp., and *Enterobacter* sp., while among the nonlactose fermenters *Acinetobacter* sp. was comparatively more common than *Pseudomonas* sp. These results are in concordance with the study done by Saad et al.[2] Among the Gram-positive bacteria, *S. aureus* was more commonly isolated than *E. faecalis*, compared with the study by Shree et al.[13]

In the present study, antimicrobial susceptibility data obtained showed that most of the Gram-negative bacilli were multidrug resistant to the routinely used antibiotics, which is an alarming situation. A high rate of antimicrobial resistance to third-generation cephalosporins was also observed. Combination antibiotics like amoxicillin/clavulanic acid also showed 61.29% sensitivity to *E. coli*, 47.36% to *Klebsiella* sp., and 20% to *Proteus* sp., while piperacillin/tazobactam showed 89.09% to *E. coli*, and 63.15% to *Klebsiella* sp. These findings are similar to the study done by Saad et al.[2] Better than best susceptibility was shown by imipenem except with *Acinetobacter* sp. Studies by Hawser et al[12] and Oteo et al[14] also showed similar findings. However, in our study, no resistance toward colistin was observed.

The rate of MRSA was 36.84%, which is in comparison with the study by Shree et al[13] and Datta et al.[15] The most active agents against MRSA were vancomycin, linezolid, quinupristin/dalfopristin, and tigecycline that showed 100% susceptibility. Also, MSSA showed 100% sensitivity to vancomycin and linezolid. Our observations are in concordance with the studies done by Datta et al[15] and Montravers et al.[16]

In *E. faecalis*, increased resistance to various antimicrobials was observed as reported by Akhter et al[17] and Chaudhary et al.[18] In our study, 37.5% showed high-level resistance to gentamicin and/or streptomycin. Combined resistance to both the high-level aminoglycosides, viz., gentamicin and streptomycin, was seen in 25% of cases. Three isolates showing high-level resistance to aminoglycosides also showed resistance to vancomycin. Our study results are in comparison to the study done by Huidrom et al.[19] But all the vancomycin-resistant *E. faecalis* showed 100% susceptibility to linezolid and tigecycline. These findings are in agreement with previous studies.[20-22]

## CONCLUSION

Cautious use of antibiotics for empirical therapy followed by modification of treatment after receiving antimicrobial susceptibility report can significantly reduce the morbidity and the mortality related to IAIs.

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