Isolation, Identification, and Antimicrobial Susceptibility Testing of Bacterial Pathogens at Chronic Sore Throat Patients Attending Outpatient Department of a Tertiary Care Hospital


Abstract

Abstract: Sore throat manifested by throat pain is a troublesome issue or discomfort for which physicians are used to prescribing antibiotics, but most often, it is found that the patient is not responding to the drug, which gives rise to the question of drug-resistant or whether there is any infection at all? Nonspecific throat pain also imparts serious public health problems. Self-medication, inadvertent and inadequate doses of antibiotics frequently prescribed by both village doctors and registered MBBS doctors is really a public health problem in developing countries like Bangladesh.

Aims: The aims of this study are to isolate the common organisms in chronic throat infection and to find out the effectiveness of various commonly used antibiotics.

Methods: The samples were collected randomly from sore throat patients via a throat swab in 5 ml of 1% sterilized saline water, and the sample was brought to the laboratory and was inoculated within two hours in both blood agar and nutrient agar (NA) media and samples were incubated at 37°C overnight. Then pure colonies were transferred to Mueller Hinton agar, and antimicrobial susceptibility testing was done by the Kirby Bauer disc diffusion method.

Results: A total of 27(24%) out of 111 samples yield positive growth. In a large number of samples, 84 (76%) did not show any isolation of the pathogenic organism. The predominant organism is a Staphylococcus aureus (55.55%). Thereafter, streptococcus (18.51%), Klebsiella pneumoniae (14.81%) and Pseudomonas (11.11%). Amoxiclav is the most resistant drug, along with cefuroxime, azithromycin, and doxycycline, respectively. Levofloxacin and ciprofloxacin show intermediate sensitivity. The most sensitive drug used in chronic sore throat found is linezolid though not frequently used, and other sensitive drugs are moxifloxacin and ceftriaxone.

Conclusion: Throat pain or soreness does not always indicate that patient has been infected by bacteria. Rationale and ethical prescription, as well as microbiological sensitivity testing, are considered to avoid drug-resistant strains.

Keywords: Sore throat, antibiotics, drug sensitivity.

Introduction

Sore throat is one of the most common indicative reasons for seeking attention from a physician. In the United States, 2.1% of ambulatory visits are due to sore throat.1 On the other hand, in developing countries, the incidence is about 7.5%. Maximum sore throats are because by upper
respiratory tract viruses, for example, rhinovirus, coronavirus, and adenovirus. The leading bacterial source of sore throat is group A β-hemolytic streptococci (GABHS), which is found in 15% to 36% of children and 5% to 17% of adults with a sore throat.

Throat pain is a common complaint to ENT specialists. Chronic or recurrent throat pain is an illness that is caused by infection of the sinuses, nasopharynx, tonsils, pharynx, and larynx, even the upper part of the esophagus. So sore throat, tonsillitis, pharyngitis, and laryngitis are commonly caused by bacterial infection. Pain is a troublesome issue or discomfort for which patients seek advice. It is common practice for physician are used to prescribe antibiotic injudiciously, but most often, it is found that the patient does not respond, which give rise to the question of drug resistance or whether there is any infection at all? However, allergic pharyngitis and nonspecific throat pain also impart serious public health problems. Self-medication, inadvertent and inadequate doses of antibiotics prescribed by unregistered village doctors is really a burning problem in developing countries like Bangladesh. Even with a registered specialist, random choice of antibiotics and lack of proper patient counseling plays an important role in developing multi-drug-resistant organisms.

Bacterial resistance to antibiotics in infection processes has been increasing in recent years and has become a serious public health problem. According to the World Health Organization (WHO), bacterial resistance to antibiotics is one of the main health problems worldwide, and no novel antibiotic has been discovered since 1987. Antibiotic resistance prolongs patients' hospital length of stay, increases the costs of treatment, and increases mortality related to infectious diseases. According to the WHO, inappropriate antibiotic use is considered the main reason for the generation of bacterial resistance to antibiotics. The abusive and indiscriminate use of antibiotics on a global scale has led to a growing concern in all health area sectors. As a result, we are on the verge of experiencing a new era where banal infections can no longer be treated with antibiotics.

**Methodology**

This prospective, observational study was conducted in collaboration with one of the specialized microbiology lab and Ear, Nose & Throat outpatient centers in Kushtia Medical College Hospital, Bangladesh, during the period from Jan 2020 to Dec 2020.

**Materials and Methods**

Specimens were collected by a trained medical technologist in the microbiology department. A sterile cotton-wool swab was rubbed over each tonsil, over the back wall of the pharynx, and over any other inflamed area. Two swabs were taken from the same areas. One was used to prepare a smear, while the other was placed into a plastic container. The specimens were processed within two hours. Smear by gram staining was done. The specimens were inoculated on Nutrient Agar, Blood Agar, and MacConkey's Agar media. Identification of the growth was made by colony morphology, gram staining, and relevant biochemical tests. Antimicrobial susceptibility testing of the isolates was performed by Kirby-Bauer modified disc-diffusion technique using Mueller Hinton agar following Clinical and Laboratory Standards Institute (CLSI) 2020 guideline with commercial discs (Oxoid, UK). The antibiotic discs used in antibiogram were amikacin (10 µg), amoxicillin-clavulanic acid (20/10 µg), ampicillin (10 µg), azithromycin (15 µg), cefepime (30 µg) cefixime (30 µg), cefoxitin (30 µg), ceftazidime (30 µg), ceftriaxone (30 µg), cefuroxime (30 µg), ciprofloxacin (5 µg), clindamycin (2 µg), cotrimoxazole (25 µg), doxycycline (30 µg), gentamicin (10 µg), levofloxacin (5 µg), linezolid (30 µg), meropenem (10 µg), moxifloxacin (5 µg), penicillin (10 unit), piperacillin-tazobactam (100/10 µg).
Results

A total of 111, with 56 female and 55 male cases of chronic throat pain not responding to antibiotics, were taken into this study. The culture and sensitivity test was done in all positive cases. Out of the 27 (24.3%), cases show positive results. A large number of patients, 84 (75.6%), did not show any growth. Among the positive group, 18 were female, and 09 were male, and the mean age was between 26 to 45 years. The most common organism isolated is *Staphylococcus aureus* (55.55%) though the primary pathogen of pharyngitis is *Streptococcus Pyogenes*. The result shows *Streptococcus* (18.51%), *Klebsiella pneumonia* (14.81%), and *Pseudomonas* (11.11%).

![Culture and sensitivity](image)

Table 1: Drug history before culture and sensitivity.

<table>
<thead>
<tr>
<th>The pattern of drug intake</th>
<th>No. of the patient (n=111)</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Regular intake</td>
<td>25</td>
<td>22.52%</td>
</tr>
<tr>
<td>Irregular intake of the drug</td>
<td>26</td>
<td>23.42%</td>
</tr>
<tr>
<td>Self-medication of antibiotic/ drug abuse</td>
<td>30</td>
<td>27.02%</td>
</tr>
</tbody>
</table>

Table 2: Organism isolated (n=27)

<table>
<thead>
<tr>
<th>Name of organism</th>
<th>No of patient</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. aureus</td>
<td>15</td>
<td>55.55%</td>
</tr>
<tr>
<td>Streptococci</td>
<td>05</td>
<td>18.51%</td>
</tr>
<tr>
<td>Klebsiellapneumone</td>
<td>04</td>
<td>14.81%</td>
</tr>
<tr>
<td>Pseudomonas</td>
<td>03</td>
<td>11.11%</td>
</tr>
</tbody>
</table>
Results & Discussion

Observational studies conducted in the UK have exposed that a huge number of patients with sore throats require to be treated with antibiotics to inhibit a single complication of infection. Evidences have revealed that one in five patients taking broad-spectrum antibiotics and one in twelve taking narrow-spectrum antibiotics suffer side effects, for example, a rash or gastrointestinal trouble, at the same time, as observational studies with routine data have connected antibiotic use straight to drug resistance at international and local levels.

In this study, the culture and sensitivity test shows that 84(75.67%) patients did not have any growth of the organism. Robinson AC et al., in their series, also show a large number none grown pathogenic organisms in their study. Study shows in both culture-positive and culture-negative group, females are more affected than male. Study Moirangthem et al. were culture positive in males is 48.78% and females is 51.36%. In our study Streptococcus pyogenes 5 (18.51%), Klebsiella pneumoniae 4 (14.81%) and pseudomonas 3(11.11%).

Regarding age & sex distribution, those who are above 26 years are more susceptible to throat infection. Though usually, children are more
susceptible to throat infections. Females (66.66%) are more victims of bacterial infection than males (33.33%), but opposite to Muhammad Ali et al. where the male is 57.2% & female 42.2%. No male & female difference by Sobhan Nandi et al. Female of the lower-class group are frequently affected may be a lack of personal hygiene. On the other hand, a study shows that the incidence of non-infective throat pain is higher in males than females.

For evaluation of the factors regarding drug-resistant history, the pattern of drug consumption by patients was taken and showed that 25(29.76%) have a regular intake of antibiotics, 26(23.42%) have irregular intake, and 30(35.71%) have a history of self-medication. Moreover, frequent use of broad-spectrum antibiotics leaves narrow-spectrum, giving rise to the development of drug-resistant. (Rewrite)

Information from Survey shows that commonly used drugs in throat pain are cefuroxime, azithromycin, amoxiclav, and doxycycline. The result of this study shows that Amoxyclav is the most resistant drug, followed by azithromycin cefuroxime and doxycycline, respectively. Levofoxacin and ciprofloxacin are still less resistant drugs. The most sensitive drug used in chronic throat pain is linezolid though it is not regularly used, and the second sensitive drug is ceftriaxone and moxifloxacin. In upper respiratory tract infection, the commonly chosen drug is amoxiclav, which shows the highest resistance level. The second popular drug preferred by the physician is cefuroxime axetil which also showed moderate resistance.

If we analyze the data, the isolated organism study shows staphylococcus is resistant to Amoxyclav11(73.33%), azithromycin 10(66.66%), cefuroxime 9(60.00%), doxycycline 5(33.33%) respectively. Highest sensitivity performed by linezolid 22(100%), Moxifloxacin 12(81.81%), doxycycline 10(66.66%). The lowest sensitivity is seen by Amoxyclav 04(26.66%) and azithromycin 5(33.33%). Levofoxacin 19(60.00%) and ciprofloxacin 19(60.00%) are still less resistant drugs.

Regarding strepto.pyogen, cefuroxime (80%), amoxyclavv3(60%), azithromycin 3(60%), resistant. Whereas ceftriaxon 5(100%), Moxifl oxacin 5(100%), linezolid 5(100%), Levofoxacin 4(80%) and ciprofloxacin 4(80%) sensitive.

Staphylococci are known to produce multidrug-resistant as it has the ability to produce penicillins, which is usually determined by the presence of the plasmid. Whereas streptococcus is a delicate organism but resistant to erythromycin Borbeau E et al. s (1982), Finland et al. 6(1976) and Seppala H. et al. v (1992) and azithromycin.

Out of four Klebsiella species, 3(75%) show sensitivity to ciprofloxacin same for levofloxacin 3(75%) and two species (50%) for cefuroxime axetil, doxycycline, and ceftriaxone 3(75%) closely matches with a study by Muhammad Ali et al. where Klebsiella showed high susceptibility to levofloxacin, Ciprofloxacin (83.3%) each, Gentamicin, Ofloxacin and Ceftriaxone (75%) each and relatively sensitive to Tobramycin (66.7%).

For 3(11.11%) Pseudomonasaureginosa Cipro and levofloxacin and moxifloxacin show 100% sensitivity supported by P.T.Wakode et al., whereas ceftriaxone show 1 (33.33%) sensitivity. This organism shows resistance to cefuroxime, azithromycin amoxiclav doxycycline. In this study, all of the organisms show resistance to more than one antibiotic, except no organism found resistant to linezolid.

Antibiogram also reveals the sensitivity of various drugs by analyzing the disc diffusion assay, where the degree of inhibition at a specific concentration (1mg/ml) of a particular antibiotic against the test organism is done. Organism shows the highest level of inhibition with linezolid and second inhibition with ceftriaxone.

Antibiotic resistance has the potential to affect people at any stage of life, as well as the healthcare, veterinary, and agriculture industries, making it one of the world's most urgent public health problems. The widespread use of antibiotics kills the susceptible cells of microbial organisms, leaving the rare resistant species to survive and
repopulate the ranks. Using an antimicrobial drug for any infection, in any dose, and over any period of time forces microbes either to adapt or to die; microbes that adapt carry genes for drug resistance that are then passed on. When an antimicrobial is not used appropriately, for the correct period of time, correct dose, at adequate potency, or for the right disease, microbes are more likely to develop resistance to that drug.

**Conclusion**

Non-infective throat pain or sore throat is not infrequent. Reasons for exploring non-infective sore throat is not the aim of this study due to lack of financial funds. Here result from this study inform the clinicians' current trends of effective antibiotics for sore throat and help them to choose proper antibiotics in the proper dose. Screening before prescribing antibiotics is also important. The selection of appropriate antibiotics, in turn, reduces the risks of antibiotic resistance. Microbiological sensitivity and resistance tests must be considered in order to avoid drug overuse and the development of drug-resistant strains.

**Conflict of interest:** None declared

**References**


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