Evaluation of the effect of some antibiotics on amoxicillin resistant bacteria isolated from middle ear infection: A comparative study

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Abstract:

Background: otitis media is one of the leading causes to loss of hears if left untreated. Its fundamental antibiotic is amoxicillin that is widely exposed to bacterial resistance. Alternatives such as cephalosporins have been become mandatory. **Objects: firstly** is to define the predominant bacteria that isolated from ear discharges. **Secondly** and most importantly is to evaluate susceptibility of bacterial isolates to each of amoxicillin, cefotaxime, ceftriaxone, and cefixime. **Thirdly** is to compare bacterial response to above antibiotics among each other. **Methods:** this study carried out **on fifty nine** infected patients with otitis media who consulted physicians at ENT department in Al-Habboubi General Hospital in Nasiriyah city from March 2014 to June 2014. Under aseptic conditions, all bacterial culturing and susceptibility tests were done. Collected data were entered into SPSS 19 and translated as o table and figures. **Results:** a total of 59 patients with a mean age of 25.53 ± 16.195 years were included in this study. It showed that *Pseudomonas aeruginosa* was a predominant *(42.4%)* followed by *Staphylococcus aureus* (37.3%). The susceptibility of *Pseudomonas aeruginosa* was the highest (40%) to amoxicillin and cefixime among whole used antibiotics. *Staphylococcus aureus* was highly sensitive to cefixime by 90.9% but amoxicillin revealed a lowest sensitivity (13.6%). Half of *E. coli isolates witnessed sensitivity by* both of cefotaxime sodium and cefixime. Enterobacter spp. isolates were completely sensitive (100%) to cefotaxime sodium and ceftriaxone. Lastly Proteus spp. was susceptible by all studied antibiotics. Cefixime was the most effective antimicrobial agent with susceptibility of 61% while amoxicillin had the lowest effectiveness (24%). **Conclusion**: The need to search new antibiotic became urgent due to prevalence of amoxicillin resistant bacteria.

Key words: Amoxicillin resistance, cefixime, cefotaxime, ceftriaxone, otitis media, susceptibility.

Introduction:

Otitis media is an inflammation in the lining of mucosa of the middle ear, causing fever, irritability, pain and other problems. Its greater causative pathogens are bacterial isolates[1]. Ineffective or belated treatment may resulting in serious complications such as hearing loss[2]. According to the estimation of World Health Organization (WHO) in 2012, more than 5% of the world's inhabitants (thirty two million children and three hundred and twenty eight million adults) have hearing loss[3]. Moreover, this disease considered a high socioeconomic load as both the urgent cause of consuming of the patient's working time and the most common reason for antibacterial medication prescription [4, 5, 6].

Amoxicillin is the intrinsic choice in otitis media remediation [7,8 ,9]. Presently, a reduction in the dosing frequency to once or twice doses per day is being prescribed, in preference to the classical three times daily doses due to ease of compliance.

Unfortunately, bacterial resistance to antibiotic is distinguished as one of the paramount threats to individual health worldwide[10]. Both of bacteriologic and clinical failures in otitis media infected people due to isolates that are non-susceptible to antibiotic has been reported previously[11]. Consequently, prevalence of penicillin resistant *pathogens*, need either to prescribe cephalosporins, or other antibacterial drugs like macrolides to be taken in the consideration as alternatives [12].

Ceftriaxone, cefotaxime, and cefixime are belonging to broad spectrum antibacterial activity, third generation cephalosporins. Ceftriaxone application is recommended in case of bacterial resistance to amoxicillin or in case of intense vomiting [13,14,12,15].

Cefixime has activity against the greatest pathogens that cause inflammation of middle ear, some performance against β-lactamase creating bacteria[16] with a few against penicillin-resistant *S.* pneumoniae. Its bioavailability is ranging between 40% and 52% after oral administration.

Cefotaxime has been used commonly to treat upper respiratory tract infections and bacterial meningitis [17]. It has less than 70% metabolism[18] and it almost excreted (40–60) % in urine as an unchanged.

Interestingly, there is a group of pathogens had been prescribed as serious cause of antibacterial resistance crisis[19,20] and termed as ESKAPE referring to first letters of each of the following bacterial names:Enterococcus, Staphylococcus, Klebsiella, Acinetobacter, *a*nd Pseudomonas, but the final E indicates both E. coli and Enterobacter. Thus, diseases resulting

from the ESKAPE organisms, in addition to other pathogens which share the same powerful of resistance, result in immense health care consumption, morbidity and mortality in the everywhere of the world [21,22,23,24,25].

From one hand, many organisms that have highly resistance to antimicrobial medications threat us at risk not only what is associated with health care but also at level of bioterrorism because it could impact on nation's security. From the other hand, despite disease prevalence, there is little evidence about the determination of the best cephalosporine should be follow amoxicillin in case of its bacterial resistance for prevention of hearing loss complication of otitis media. These reasons leading us to design this study which aims to: indicate the predominant bacterial isolates from middle ear infected persons, evaluate the susceptibility of each of amoxicillin, cefotaxime, ceftriaxone, and cefixime by isolates, as well as to assess extent of reduction of bacterial infection using these cephalosporin antibiotics in comparison to that of amoxicillin as a typical treatment.

Materials and Methods

colle Data

Ction and sampling:

This cross section study was conducted on 59 patients with otitis media with effusion as diagnosed clinically. The samples were collected at ENT Department at Al-Habboubi General Hospital in Nasiriyah city, from March 2014 to June 2014. Samples gathering had been carried out using sterile swab sticks which were labeled for each patient. After collection, these sticks were taken instantly to the Microbiology Laboratory of the Hospital for culturing.

Laboratory Diagnostic Methods

Sterilized inoculation of specimens was applied onto chocolate, MacConkey, and blood agar plates. All of these plates were set in an aerobic incubator while incubation of chocolate plate has been done in a CO_2 reinforced atmosphere (candle jar) at 37°C and for 24 hours.

Whole isolates were examined for their susceptibilities which achieved on Mueller-Hinton agar utilizing adjusted Kirby-Bauer disc diffusion mode[26]. The susceptibility manner for each of bacterial isolate was translated according to the specific scales of Clinical and Laboratory Standards Institute[27]. Tests of bacterial sensitivity were accomplished for each of: amoxicillin, cefotaxime, ceftriaxone, and cefixime.

Statistical Analysis:

The data included in this study were analyzed statistically using SPSS (version 19) software. Both of non-parametric chi-square (χ^2) test and T-tests were applied to measure association among groups.

Results:

The data of this prospective, randomized trial had been conducted through the period (March 2014 to June 2014). A total of 59 patients diagnosed with infection of otitis media were included in the study and from them bacterial isolates were obtained. The age mean of patients was 25.53 ± 16.195 years. There was non-significant difference in distribution between female (n=36) 61% and of male (n=23) 39% with P-value > 0.05.

The predominant bacterial species isolated were Pseudomonas aeruginosa (42.4%) followed by Staphylococcus aureus (37.3%), Proteus spp. (13.6%) and both of E. coli and Enterobacter spp. which had the same frequency (3.4%) as presented in figure 1.

Figure 2 with its attachment table showed a comparison of the susceptibilities among the four antibacterial agents by different bacterial isolates. It is very clear that amoxicillin faced wide range of resistance by studied organisms versus cephalosporin group.

When amoxicillin had been examined, it found that the highest susceptibility was by Pseudomonas aeruginosa (only 40%), the next susceptibilities were via both Staphylococcus aureus and Proteus spp. which were approximately similar 13.6% and 12.5% respectively. Unluckily, it faced 100% resistance by each of E. coli and Enterobacter spp.

In contrast, ceftriaxone encountered complete susceptibility by Enterobacter spp. (100%), then (81.8%) by Staphylococcus aureus. (75%) by Proteus spp., and 36% by Pseudomonas aeruginosa but there was no any response by E. coli.

Also, cefotaxime sodium witnessed (100%) susceptibility by Enterobacter spp. Followed by 75% and 72.7% by each of Proteus spp. and Staphylococcus aureus respectively. Half of E. coli isolates (50%) and only (32%) of Pseudomonas aeruginosa isolates were susceptible.

Lastly, cefixime had been showed that the major susceptibility was represented by Staphylococcus aureus isolates (90.9%). It was susceptible by just half (50%) by each of Enterobacter spp., Proteus spp. and E. coli. isolates. Less susceptibility was observed by Pseudomonas aeruginosa (40%).

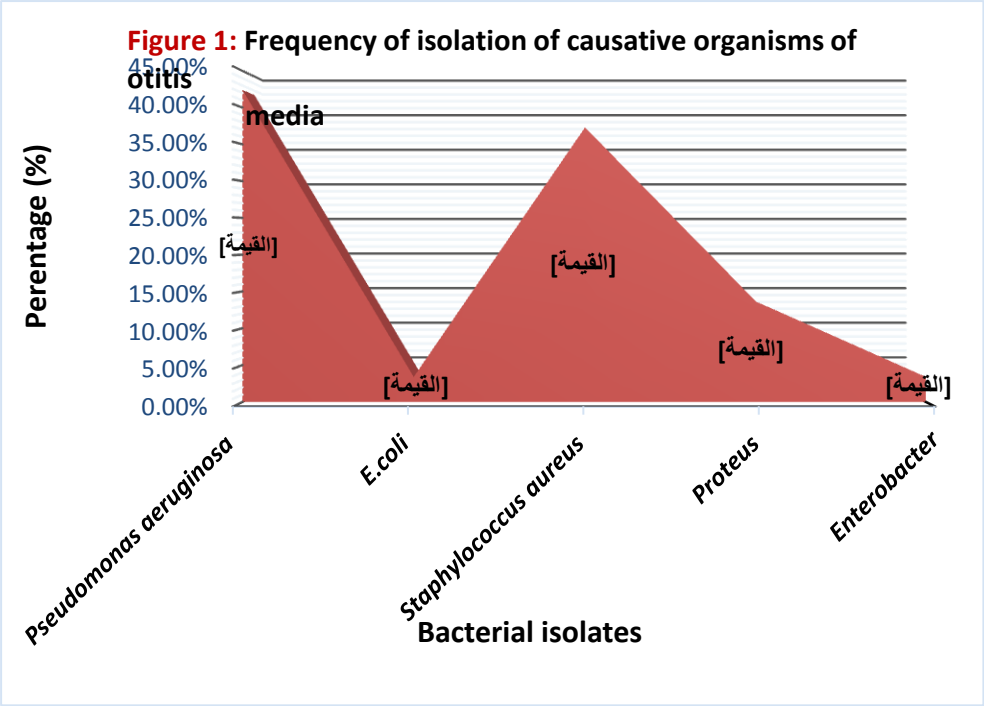
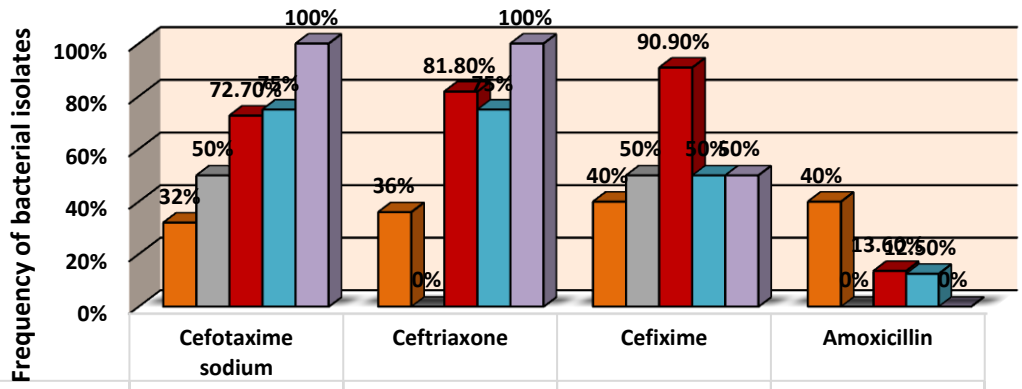


Figure 2: Comparison of the antibacterial susceptibility of amoxicillin, cefotaxime sodiume, ceftriaxone and cefixime by different bacterial isolates



	Cefotaxime sodium	Ceftriaxone	Cefixime	Amoxicillin
Pseudomonas aeruginosa	32%	36%	40%	40%
E.coli	50%	0%	50%	0%
Staphylococcus aureus	72.70%	81.80%	90.90%	13.60%
Proteus	75%	75%	50%	12.50%
Enterobacter	100%	100%	50%	0%

Pseudomonas aeruginosa E.coli Staphylococcus aureus Proteus Enterobacter

Overall, cefixime was the most effective antimicrobial agent with a susceptibility of 61%, followed by ceftriaxone (59%) and cefotaxime sodium (56%), whereas amoxicillin showed the lowest response (24%). (Figure 3).

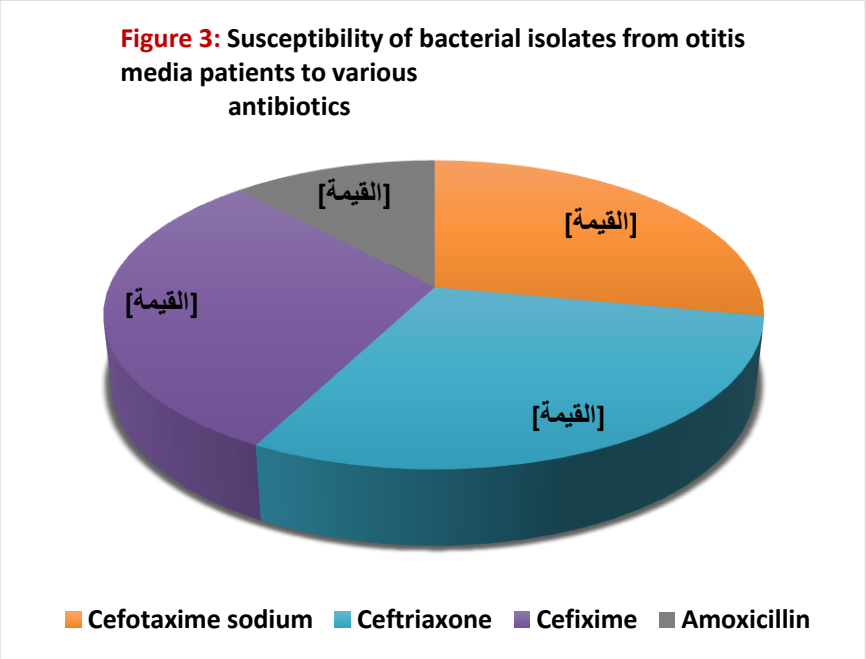


Figure 3: Susceptibility of bacterial isolates from otitis media patients to various antibiotics

Discussion:

Otitis media infection, in particular that associated with discharge is a leading cause of a serious hearing loss complication[2] if it does not exposed to an appropriate therapy. The disease medications stay largely empirical, since diagnostic procedures and microbiological techniques that needed for culture are often costly over and above it may be considered as a time consuming.

In the present study, the incidence of otitis media with effusion was non-significantly higher in female than that in male (P value > 0.05) which is similar to that of published studies [28, 29].

In accordance to outcomes of some previous studies [30, 29], this study revealed similarity in pattern of middle ear bacterial predominance, where the most common otitis media pathogen was P. aeruginosa followed by *S. aureus*. (Figure1).

The findings in figure 2 highlighted that amoxicillin (which had been considered the mainstream remedy for bacterial infections of otitis media) could be eradicate only beneath the half of isolates (40%) and should be taken in to account that this represents the highest bacterial susceptibility to amoxicillin among all different studied pathogens. This highest susceptibility was against the most prevalent bacteria P. aeruginosa. The study results disclosed another drawback about amoxicillin which *is* represented by its susceptibility by the second predominant pathogen *S.* aureus that

was around only 13.6%, it is close to Proteus spp. susceptibility. Furthermore, absence of response by both *E.* coli and Enterobacter spp. means complete resistance to amoxicillin by these organisms. The findings of this study found consistence to Italian study where amoxicillin witnessed bacterial resistance by about 81.9% of isolates[31]. This resistance interpretation may be due to broad spectrum of amoxicillin activity in addition to its overuse. Alternative strategies like utilizing cephalosporin therapy will become key elements. In current study, three antibiotics from cephalosporin family were studied. Ceftriaxone is one of them, when it analyzed, the outcome has been shown that sensitivity against each of *S. aureus* and Proteus spp. is three folds higher in ceftriaxone in comparison to amoxicillin. In spite of the similarity between these two antibiotics against *P. aeruginosa* and equality against E. coli but there was inverse relationship between each other with Enterobacter spp. where complete sensitivity has been observed with ceftiaxone which is absolutely different from that of amoxicillin. These outcomes are contracted to that of Argaw-Denboba et al study in which ceftriaxone faced a great resistance (84.5%)[31]. This difference in ceftriaxone findings between two studies may be due to the difference in how many ceftriaxone has been prescribed. In our country this medicine is not prescribed widely for otitis media patients.

From the same figure, cefotaxime sodium results existed wider range of its antibacterial activity in comparison to amoxicillin except P. aeruginosa which reflected approximately similar activity to both cefotaxime sodium and to amoxicillin (that is less than the half of the whole isolates). This result is inconsistence with a study published recently [29] which reported resistance of P. aeruginosa to cefotaxime, which can interpreted by wide prescription or dealing with this medication, the reason behind this resistance.

Findings of cefixime antibacterial susceptibility has been revealed its highest susceptibility against S. aureus (90.9%), as well as broader impact versus all studied pathogens than amoxicillin, while Johnson et al concluded that cefixime has equivalent bacteriologic efficacy to amoxicillin [32] this disagreement may belong to appropriate prescription of cefixime to a suitable disease, making no chance for bacteria to change antibacterial target or key sites like bacterial enzymes or antibacterial permeability. Consequently, cefixime activity will increase.

Differences in the bacterial susceptibility to amoxicillin, ceftriaxone, cefotaxime sodium, and cefixime exist clearly. Focusing on the effect of the four medications on predominant organisms causing otitis infection *(*P. aeruginosa and S. aureus), cefixime has been achieved the highest effectiveness in comparison to others, while amoxicillin had minimum effect.

Knowing of the best antibacterial choice for middle ear infected patients rely on comparison the sensitivity of all types of

isolated bacteria from patients' middle ear against studied antibiotics. Figure 3 illustrated that cefixime has greatest activity (61%), followed by ceftriaxone and cefotaxime sodium (59%) and (56%) respectively. The probable reason for this outcome is due to low drugs consumption. Surprisingly, amoxicillin has the lesser susceptibility by bacterial isolates (24%). Thus, it may be considering cefixime as a first line treatment for otitis media infection followed by either ceftriaxone or cefotaxime sodium.

Conclusions:

From this study, it can be concluded that otitis media is still one of the most common problems threatening ear health and it is more prevalent among females than males. The predominance is to Pseudomonas aeruginosa pathogens which for it both of amoxicillin and cefixime are susceptible by only 40% (the major susceptibility in this study toward this organism). This pays us to search new antibacterial agents for acute otitis media Pseudomonas aeruginosa. In general, cefixime is the most effective antibiotic for inhibition almost all types of otitis media bacteria. Ceftriaxone and cefotaxime sodium may be considered as the second choice in relation to bacterial susceptibility. Amoxicillin is preferred to be last choice in treatment of patients with otitis media due to prevalence of resistance by many of pathogens that infect middle ear.

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تقييم تأثير بعض المضادات الحيوية على البكتيريا المقاومة للأموكسيسيلين والمعزولة عن عدوى الأذن الوسطى: دراسة مقارنة

عبير مظفر عبد الرحمن

ماجستير في علم الأدوية/كلية الصيدلة/جامعة ذي قار

الخلاصة:

يعد التهاب الأذن الوسطى أحد الأسباب المؤدية لفقدان حاسة السمع إذا ترك بدون علاج. المضاد الحيوي الأساسي لهذا المرض هو أموكسيسيلين الذي تعرض بشكل واسع للمقاومة البكتيرية. البدائل مثل سيفالوسبورينز أصبحت ضرورية. الأهداف: أولاً هو التعرف على البكتريا المهيمنة المعزولة من السوائل المتدفقة من الإذن. ثانياً والأكثر أهمية هو تقييم حساسية العزلات البكتيرية لكل من أموكسيسلين، سيفوتاكسيم، سفترايكسون، والسفيكسيم. ثالثاً هو مقارنة الاستجابة البكتيرية للمضادات الحيوية أعلاه بين بعضهم البعض. طريقة العمل: أجريت هذه الدراسة على ٥٩ مريضاً مصابا ب التهاب الأذن الوسطى والذين استشاروا الأطباء في قسم الأنف، الأذن، والحنجرة في مستشفى الحبوبي العام في مدينة الناصرية من آذار ٢٠١٤ إلى حزيران ٢٠١٤. تحت ظروف معقمة، تم إنجاز كل الأوساط الزراعية واختبارات الحساسية. البيانات التي تم جمعها ادخلت إلى لبرنامج الإحصائي ١٩ ومن ثم ترجمت إلى جدول وفقرات. النتائج: ٥٩ مريضاً مع متوسط عمر٢٥.٥٣ ± ١٦.١٩٥ سنوات تم تضمينهم في الدراسة. وقد تبين أن الزائفة الزنجارية كانت هي السائدة (٤٢.٤%) تليها المكورات العنقودية الذهبية (٣٧.٣%). حساسية الزائفة الزنجارية كانت الأعلى (٤٠%) على أموكسيسلين وسفيكسيم بين كل المضادات الحيوية المستخدمة. المكورات العنقودية الذهبية كانت عالية الحساسة وبنسبة ٩٠.٩% ل سفيكسيم، ولكن النتائج كشفت أن أقل حساسية ل لمكورات العنقودية الذهبية تعود ل أموكسيسلين (١٣.٦%). نصف عزلات الأشريكية القولونية شهدت حساسية من قبل كل من سيفوتاكسيم صوديوم وسفيكسيم. عزلات المكورات المعدية كانت كاملة الحساسية (١٠٠%) ل سيفوتاكسيم صوديوم وسفترايكسون. أخيراً البكتريا المتقلبة كانت حساسة لكل المضادات الحيوية التي شملتها الدراسة. هذه الدراسة كشفت أن سفيكسيم لديه أكبر فعالية مضادة للبكتريا بحساسية ٦١% بينما أموكسيسلين لدية أقل فعالية ٢٤%. الاستنتاجات: الحاجة لبحث عن مضاد حيوي جديد أصبح ملحاً بسبب انتشار البكتريا المقاومة ل أموكسيسيلين.