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Aerobic Bacteria and Antibiotic Sensitivity on Odontectomy Wound in RSUD Arifin Achmad Riau

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ABSTRACT

Background and aim: The impacted tooth is defined as a condition where the tooth fails to erupt into the dental arch. It could be extracted with a procedure called odontectomy. The most common complication of this procedure is infection by aerobic as well as anaerobic bacteria, and antibiotics could treat this complication. This study aimed to identify the type of bacteria found on the wound of odontectomy patients and the antibiotics sensitivity pattern in RSUD Arifin Achmad Riau.

Material and methods: Patient specimens after odontectomy were collected from wound swabs. All specimens were then cultured and identified for the type of bacteria, and an antibiotic sensitivity test was done by Kirby-Bauer method according to the Clinical and Laboratory Standard Institute (CLSI).

Results: This study found that the most dominant population patient odontectomy is female (66.67%), 15-55 years old, graduated from senior high school (63.33%), the third molar as the affected tooth (86.67%), and located in the lower jaw (100%). Gram-positive bacteria (52%) followed by Alpha-hemolytic Streptococcus (40.74%), Staphylococcus aureus .22,22%) and Coagulase Negative Staphylococci (37.04%). Gram-negative bacteria (48%) followed by Klebsiella sp. (56%), Enterobacter sp. (32%), Pseudomonas sp. (8%), and Escherichia coli (4%). The antibiotics with the highest sensitivity were levofloxacin, meropenem, chloramphenicol, gentamicin, sulphamethoxazole, amikacin, ceftazidime, fosfomycin, and ciprofloxacin. All bacteria were resistant to ampicillin and metronidazole.

Conclusions: Klebsiella sp and Alpha-hemolytic Streptococcus are the most dominant bacteria found in the wound of odontectomy patients. Antibiotics recommended for post-odontectomy wounds are Carbapenem and Quinolones.

1. Introduction

An impacted tooth was a condition where a tooth fails to erupt into the dental arch at the estimated time.[1] This condition could manifest as a blunt pain on the jaw that radiates to the neck, ear, and temporal lobe (migraine) due to compression to the adjacent nerve, such as the inferior alveolar nerve by the tooth. Long-term complications include pain, dental caries, pericoronal infection, periodontitis, damage to the other teeth, and in an extreme case, could lead to an abscess, cyst, tumor, or massive damage to the jaw.[2, 3] The most affected tooth is the third molar because it was the last tooth to erupt. Approximately 25% of people only had three third molar, and 9 of 10 people experienced impacted third molar.[2, 4] The prevalence of third molar impaction is 16,7%-68,6%, with light-skinned European and Chinese Singaporeans females having a greater risk than males.[5] Other studies showed that the prevalence of third molar impaction is 16-73% in young adults.[6] In Indonesia, third molar impaction has also been challenging to treat.[7] The impacted tooth could be extracted by odontectomy. The high prevalence of tooth impaction leads to a high frequency of odontectomy procedures. In Indonesia, odontectomy has a high prevalence of 0.3%. Province of Riau has a high prevalence of this procedure (0.2%) in patients aged 35-44 years old (0.4%), female (0,3%), and having a college degree (0.8%).[7] Odontectomy was classified as a minor surgery with several complication risks, including injury to the inferior alveolar nerve, edema, surgical site infection, and iatrogenic complication.[2, 4] Other studies reported another list of complications that could arise from odontectomy, including bleeding, edema, pain, trismus, ulcer, and infection.[8, 9] Surgical site infection could have various etiological microorganisms, including bacteria. The most predominant bacteria found in the surgical wound is aerobic bacteria such as Staphylococcus, Bacillus, Haemophilus, Corynebacterium, Stenotrophomonas, Streptococcus. Anaerobic bacteria such as Fusobacterium, Peptosptreptococcus, Prophyromonas, and Prevotella strain

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are also reported as the etiology of surgical site infection.[4] Management of surgical site infection includes various treatments by antibiotics. First-line antibiotics for dental infection, including surgical site infection, were penicillin and metronidazole.[10, 11] Wide-spectrum antibiotics like all generations of cephalosporin and ward-level antibiotics such as ceftriaxone and cefadroxil also have been widely used for surgical site infection.[4] Irrational antibiotic use was a global crisis leading to antibiotic resistance, including multidrug resistance. RSUD Arifin Achmad Riau is the referral hospital of Riau province that performs odontectomy. High surgical site infection risk from odontectomy procedure results in the frequent use of antibiotics, leading to a high risk of antibiotic resistance. This study was aimed to identify the type of bacteria and antibiotic sensitivity of the bacteria on the odontectomy wound in RSUD Arifin Achmad Riau Province.

2. Material and methods

This research is a descriptive study with consecutive sampling. The research place for oral surgery is Arifin Achmad Hospital, Riau Province. Swab collection of post-odontectomy wound patients and the Microbiology laboratory is Faculty of Medicine, Universitas Riau for laboratory analysis from May to November 2021. This research has gone through an ethical review procedure and has received an ethical letter from the research ethics unit and health of the Faculty of Medicine, Riau University, with Decree number B/058/UN19.5.1.1.8/UEPKK/2021. The research sample was post-odontectomy patients at the oral surgery clinic of Arifin Achmad Hospital, Riau Province, who met the inclusion criteria, namely patients aged 15 years, types of impacted teeth in the form of canines, premolars and molars, location of maxillary and mandibular impacted teeth, came for post-odontectomy wound control. For the first time on day 5, read the study by inviting informed consent. The exclusion criteria were post-odontectomy wounds treated or patients' wounds that had healed the minimum sample size of 30 samples.[17] Specimens using a sterile swab were taken to the media Trypticase soya broth (TSB) and Thyoglycolate broth and brought to the Microbiology Laboratory, Faculty of Medicine, Universitas Riau within 2 hours for culture. TSB media and Thyoglycolate broth containing samples were incubated at 37ºC for 18-24 hours. Samples that have been incubated are taken with a loop, inoculated onto blood agar media, and Mac Conkey agar (Oxoid Ltd) incubated at 37ºC for 24 hours.

Furthermore, bacterial identification was carried out by Gram staining, colony morphology, and biochemical tests such as catalase, oxidase, coagulation, and novobiocin tests for Gram-positive and Triple Sugar Iron Agar (TSIA) test, motility test, and citrate test for Gram-negative. Antibiotic sensitivity test by disc diffusion method (Kirby Bauer procedure) with Mueller-Hinton agar (Oxoid Ltd) and various antibiotic discs were incubated at 37°C for 24 hours. The results are seen in the form of a clear bacterial zone as measured by a caliper. Sensitive, intermediate, and resistant analyzes were based on tables from the Clinical and Laboratory Standard Institute (CLSI) guidelines.[15] The study results were presented in the form of a frequency distribution table and expressed in percentages.

3. Results

Odontectomy patients characteristics

Characteristics from 30 patients were registered as study samples, and the characteristics were presented in Table 1.

Table 1. Study samples characteristics.

Characteristics	N	%
Gender		
Male	10	33.33
Female	20	66.67
Total	30	100
Age	15-55 years old	
Total	30	100
Education		
Junior High School	3	10
Senior High School	19	63.33
College	8	26.67
Total	30	100

Assessment of type and location of the impacted tooth from all odontectomy patients found 3.33% canine impaction, 10% premolar impaction, and 86.67% molar impaction. All impaction is located in the lower jaw (100%). Data were presented in Table 2.

Table 2. Type and Location of the impacted tooth.

Jenis dan Lokasi	N	%
Types		
Canine	1	3.33
Premolar	3	10
Molar	26	86.67

Total	**30**	**100**
Location		
Upper Jaw	0	0
Lower Jaw	30	100
Total	**30**	**30**

Aerobic bacterial pattern on odontectomy wound

The samples culture and identification (macroscopic and microscopic) were presented in Table 3. This study found that several samples show multiple bacterial growths. Identification from the culture shows a 52% growth of gram-positive bacteria and 48% growth of gram-negative bacteria. Gram-positive bacteria identified to consist of Alpha-hemolytic Streptococcus (40.74%), Staphylococcus aureus (22.22%), and Coagulase Negative Staphylococci (CNS) (37.04%). Gram-negative bacteria identified to consist of Klebsiella sp. (56%), Enterobacter sp. (32%), Pseudomonas sp. (8%), and Escherichia coli (4%) (Table 3).

Table 3. Aerobic bacterial pattern on odontectomy wound.

Type of Bacteria	N	%
Gram-positive Bacteria		
Alpha-hemolytic Streptococcus	11	40.74
Staphylococcus aureus	6	22.22
Coagulase Negative Staphylococci (CNS)	10	37.04
Total	27	52
Gram-negative Bacteria		
Klebsiella sp.	14	56
Enterobacter sp.	8	32
Pseudomonas sp.	2	8
Escherichia coli	1	4
Total	25	48
Total	52	100

Antibiotic sensitivity pattern in odontectomy wound

The bacteria that have been tested for antibiotic sensitivity. The antibiotic sensitivity test results against alpha-hemolytic Streptococcus were 100% sensitive to levofloxacin and meropenem, respectively, followed by ciprofloxacin (82%), gentamicin (73%), sulphamethoxazole, and tigecycline 64% each. Antibiotic resistance was found in ampicillin, clindamicin and metronidazole. (Fig. 1). Staphylococcus aureus was 100% sensitive to chloramphenicol, gentamicin, levofloxacin, and meropenem. Meanwhile, it shows 83% sensitivity to sulfamethoxazole, 66% sensitivity to cefoxitin and tigecycline, and 50% sensitivity to ciprofloxacin and clindamycin. Resistance was found for ampicillin, metronidazole, and vancomycin (Fig. 2). Klebsiella sp. shows the highest sensitivity to levofloxacin and meropenem (93%), followed by amikacin, ceftazidime, chloramphenicol, gentamicin, and sulphamethoxazole with a sensitivity percentage of 86%. Klebsiella sp. was also sensitive to fosfomycin with 79% sensitivity and ciprofloxacin and tigecycline with 72% sensitivity. Resistance was found for ampicillin and metronidazole. (Fig. 3). Enterobacter sp. was 100% sensitive to chloramphenicol, fosfomycin levofloxacin, meropenem, and sulphamethoxazole, followed by 87% sensitivity to amikacin, ceftazidime, ciprofloxacin, and gentamicin. Resistance was found for ampicillin and metronidazole. Pseudomonas sp. shows the highest sensitivity to amikacin and sulfamethoxazole with a percentage of 100%. It shows a 50% sensitivity to ceftazidime, chloramphenicol, ciprofloxacin, fosfomicin, gentamicin, levofloxacin, meropenem and metronidazole. Pseudomonas sp. shows resistance to ampicillin dan tigecycline (Fig. 4). Antibiotic sensitivity test of Escherichia coli shows sensitivity to all antibiotics except ampicillin, metronidazole, and sulfamethoxazole.

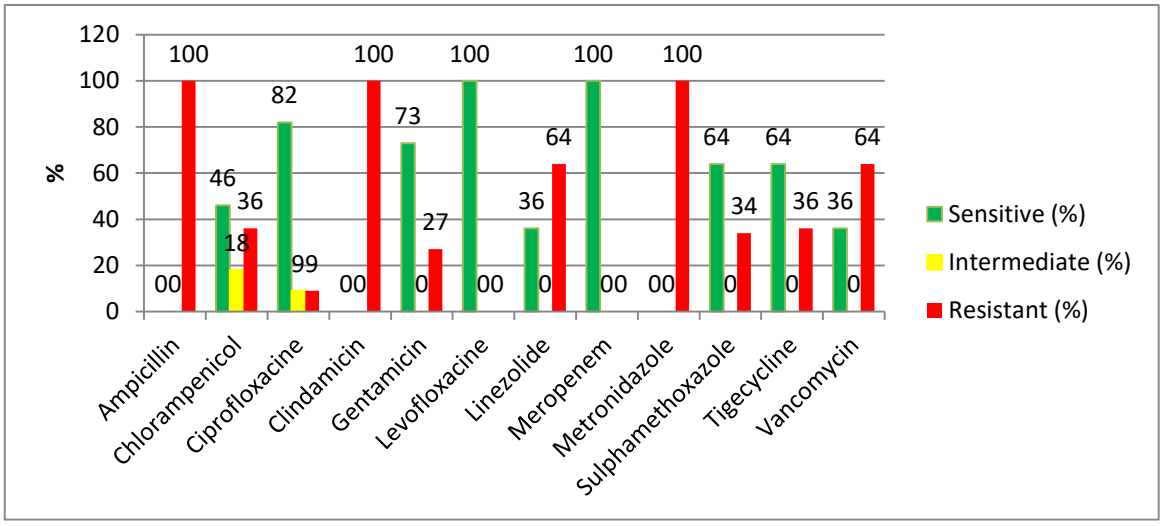

Fig. 1. Antibiotic sensitivity pattern of alpha-hemolytic streptococcus (n=11).

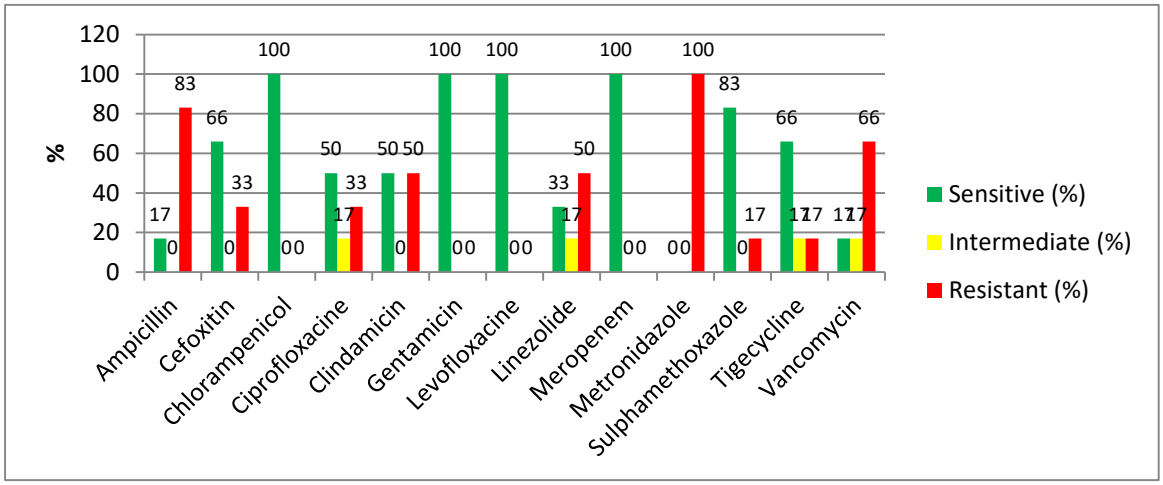

Fig. 2. Antibiotic sensitivity pattern of staphylococcus aureus (n=6).

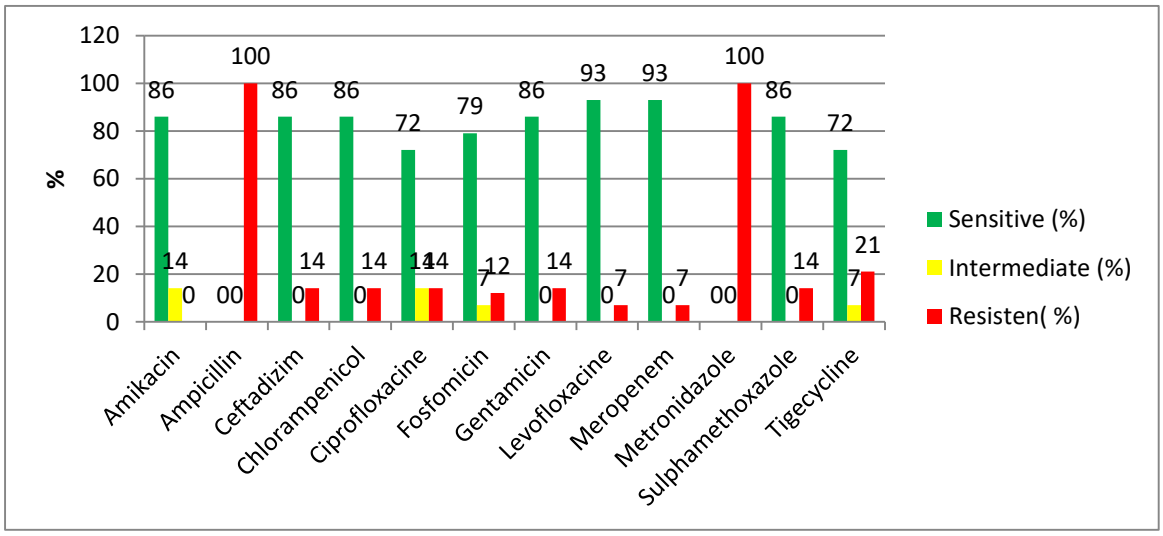

Fig. 3. Antibiotic sensitivity pattern of klebsiella sp (n=14).

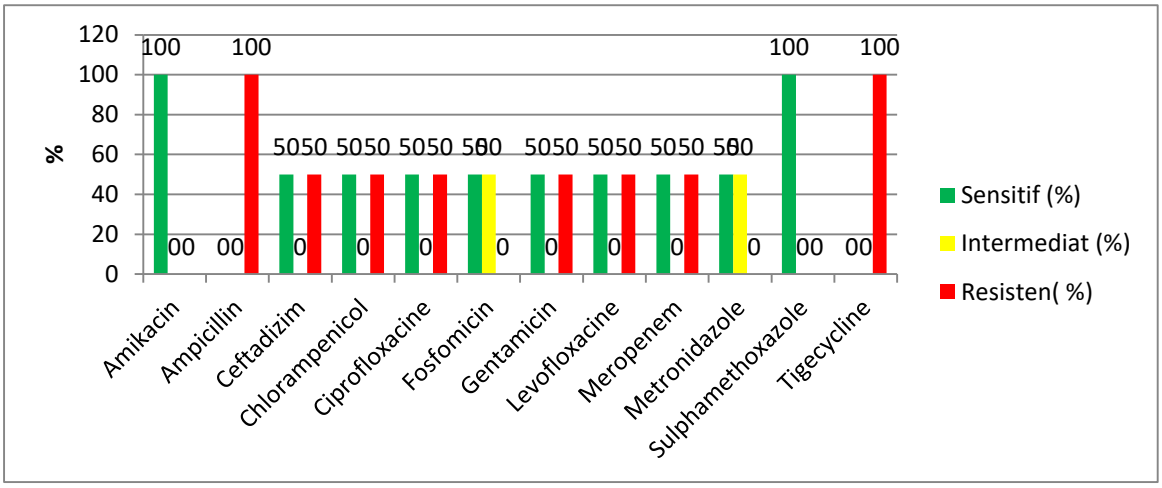

Fig. 4. Antibiotic sensitivity pattern of pseudomonas sp (n=2).

4. Discussion

Table 1 shows the characteristic of the study samples with females (66.67%) aged 15-55 years old as the most predominant. This result is in line with another study that reported odontectomy was more frequent in females (61.37%) and younger age, approximately 16-25 years old (38.7%).[3] Another study reported that odontectomy was more frequently performed in female and young adults.[7] On the other hand, a study conducted by Hasan found that odontectomy was more frequently performed in males (52.6%), but the average age is the same, which is 29-30 years old (7.63%).[5] Other study also found that odontectomy was more frequent in males (55%) in comparison with females (49%).[19] On the educational degree, the highest prevalence was found in the population with junior high school degrees (63.33%), while a report published by the Indonesian Ministry of Health showed that odontectomy was more frequent in the population with a college degree.[7] Based on Table 2, odontectomy was most frequently performed in the third molar (86.67%), and all affected tooth was located in the lower jaw (100%). The third molar is the most frequent teeth impacted, with a 16-73% prevalence.[1, 6] High prevalence of third molar impaction results from the third molar being the last erupted teeth in the upper or lower jaw. Normally, the third molar will erupt at 16-25 years old. In this age group, bone calcification has been completed, leading to the difficulty of the third molar erupting to the surface.[1] A study at Saudi reported that impacted third molars were found more frequently in the lower jaw (53.1%) than in the upper jaw (31.8%), and their extraction was carried out by odontectomy[5] Odontectomy was a minor surgery performed to extract the impacted tooth. Multiple factors could increase the risk of tooth impaction, including genetics, the absence of tooth buds, impacted tooth buds, and external factors such as nutrition. Malposition of the tooth buds or angulation (mesial, distal, vertical, and horizontal) could also result in impaction because the tooth buds will erupt in the wrong path. Deviation of the tooth buds angulation could lead to a partial eruption of the teeth (soft-tissue impacted) or no eruption at all (total/bony-impacted).[1] This study found that in the odontectomy wound, the most predominant bacteria is the gram-positive bacteria such as alpha-hemolytic Streptococcus (40.74%), Staphylococcus aureus (22.22%), and Coagulase Negative Staphylococci (CNS) (37.04%). Gram-negative bacteria such as Klebsiella sp. (56%), Enterobacter sp. (32%), Pseudomonas sp. (8%), and Escherichia coli (4%) were also found in a lower percentage (Table 3).

The finding was consistent with the study conducted by Barasa et al., that isolate Gram-positive bacteria such as Staphylococcus aureus (40%) and Streptococcus pyogenes (5%) also Gram-negative bacteria such as Pseudomonas sp. (23%), Klebsiella sp. (23%), Proteus mirabilis (11%), and Escherichia coli (2%) from the odontectomy wound.[19] Another study reported the identification of Alpha-hemolytic Streptococcus (40%), Coagulase Negative Staphylococci (CNS) (40%), and Staphylococcus aureus (20%) in the dentoalveolar abscess.[16] Endriani et al. (2020) identified 26.3% of Streptococcus sp. in dental caries.[17] According to a publication from Matsumoto (2018), the most frequent aerobic bacteria to be isolated from dentoalveolar infection are Streptococcus viridans and Streptococcus anginosus,[18] while Barasa et al. (2015) reported that the most frequent bacteria to be isolated from the orofacial area are Staphylococcus aureus, Klebsiella sp., and Pseudomonas sp.[19] This study showed that the number of Alpha-hemolytic Streptococcus was higher. These bacteria are normal flora of the oral cavity but can cause opportunistic infections, possibly due to odontectomy performed.[20] Alpha hemolytic Streptococci have high sensitivity to levofloxacin and meropenem, but resistance is found to ampicillin, clindamycin, and metronidazole (Figure 1). Figure 2 shows the antibiotic sensitivity test of Staphylococcus aureus. Staphylococcus aureus was sensitive to chloramphenicol, gentamicin, levofloxacin, and meropenem but showed resistance to ampicillin, metronidazole, and vancomycin. The study conducted by Barasa et al. (2015) found that Staphylococcus aureus shows a high antibiotic sensitivity toward vancomycin (100%), oxacillin (92%), and cefotaxime (90%), while ampicillin shows the lowest sensitivity with the percentage of 25%.[19] Ampicillin resistance in this study could be developed because isolates (33,3%) were identified as Methicillin-Resistant Staphylococcus aureus (MRSA). According to the Clinical and Laboratory Standard Institute (CLSI) guideline, an isolate that was resistant to methicillin or cefoxitin was automatically resistant to all antibiotics in the β-lactam class.[15] MRSA could develop because Staphylococcus aureus has a specific DNA element size 20-100 kb called Staphylococcal Cassette Chromosome mec (SSCmec) located in the chromosome (mecDNA). SCCmec contains mecA, a gene that encodes the transformation of PBP2 into PBP2a, which makes the receptor for β-lactam antibiotics unrecognizable. Another factor that promotes the development of resistance is the β-lactamase enzyme produced by Staphylococcus aureus.[21] Klebsiella sp. shows the highest sensitivity to levofloxacin and meropenem (93%), followed by amikacin, ceftazidime, chloramphenicol, gentamicin, and sulphamethoxazole with a sensitivity percentage of 86%. Klebsiella sp. was also sensitive to fosfomycin with 79% sensitivity and ciprofloxacin and tigecycline with 72% sensitivity

(Figure 3). Enterobacter sp. was 100% sensitive to chloramphenicol, fosfomycin levofloxacin, meropenem, and sulphamethoxazole, followed by 87% sensitivity to amikacin, ceftazidime, ciprofloxacin, and gentamicin. Klebsiella sp and Enterobacter sp were resistant to ampicillin and metronidazole. Escherichia coli species all appeared to be sensitive to the tested antibiotics except ampicillin, sulphamethoxazole, and metronidazole which were resistant. These results show that the Enterobacteriaceae bacteria are ineffective against penicillin and metronidazole groups.

The antibiotic sensitivity test result of Pseudomonas sp. represented in Figure 4 shows that Pseudomonas sp. is 100% sensitive to amikacin and sulfamethoxazole. This result is in line with the study conducted by Barasa et al. (2015) that found Pseudomonas sp. has 75% sensitivity to amikacin and 7% sensitivity to amoxiclav.[19] This study also found that Pseudomonas sp. is sensitive to levofloxacin with a sensitivity percentage of 50%, which is consistent with the study conducted by Yadav et al. (2017) found 83.2% sensitivity to levofloxacin.[22] The sensitivity of Pseudomonas sp. might be different in each medical facility due to the difference in environment and facility. The increased prevalence of resistance towards different antipseudomonal has been reported globally and has become a serious problem in treating infection caused by Pseudomonas sp.[22] This study showed that all bacteria were highly sensitive to levofloxacin and meropenem except Pseudomonas sp (50%). Levofloxacin and meropenem are broad-spectrum antibiotics active against all Gram-positive and Gram-negative bacteria except for oxacillin-resistant Staphylococcus bacteria, some Enterobacteriaceae, and Pseudomonas.[20] This study found that all bacteria isolated were resistant to ampicillin. The high resistance to ampicillin in this study may be because ampicillin is the first choice for treating infections in the oral cavity, including post-odontectomy infections, so it is often used.[10, 11] Ampicillin is a broad-spectrum penicillin antibiotic in which bacteria produce a beta-lactamase enzyme called penicillinase which can damage the beta-lactam ring by hydrolyzing penicillin into psilotic acid, which is no longer active as an antibacterial.[20] High resistance to ampicillin in this study could have resulted from the fact that ampicillin is the first-line management of dentoalveolar infection, including wound post odontectomy.[10, 11] Ampicillin is an antibiotic from the penicillin class. Antibiotics in the penicillin class have a special β-lactamase called penicillinase that could destroy the β-lactam rings by hydrolyzing penicillin into inactive penicilloic acid that acts as an antibacterial.[20] The high metronidazole resistance in this study was probably due to having insignificant activity against facultative aerobic or anaerobic bacteria.[20] Bacterial identification and antibiotic sensitivity tests conducted on the bacteria from odontectomy wounds could be used as strategies to optimize the patient's management, reduce postoperative complications, and reduce the medical expenses for the treatment post odontectomy.

Limitations of study

The limitation of this study is the small number of samples, so it cannot show the proper condition of bacterial patterns and antibiotic sensitivity. Therefore, it is hoped that the research will continue with an adequate number of samples or more.

5. Conclusions

Bacteria often found in post-odontectomy wounds are Klebsiella sp and Alpha-hemolytic Streptococcus. Antibiotics with the highest sensitivity to bacteria in post-odontectomy wounds in Gram-positive were levofloxacin, meropenem, chloramphenicol, and gentamicin, while for Gram-negative were levofloxacin, meropenem, sulphamethoxazole, amikacin, ceftazidime, levofloxacin, gentamicin, chloramphenicol, and doxycycline. Almost all bacteria are resistant to ampicillin and metronidazole. Antibiotics recommended for post-odontectomy wounds are Carbapenem and Quinolones.

Conflict of Interest

The authors declared that there is no conflict of interest.

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