Antibiotic Resistance and Biofilm Composition of E. coli, K. Pneumoniae, S.Aureus Isolated from Surgical Procedures

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Abstract
Collected (154) samples of patients with surgical inflammation reviewed and lying at Ramadi General Teaching Hospital and Women's and Children's Hospital in Ramadi (Anbar province), the process of collecting samples began from 4/10/2021 to 1/2/2022 i.e. (October 4, 2020) 21 to February 2022, the study included patients (caesarean sections, appendicitis, tonsils, colon-hemorrhoids), both sexes and of different ages, to investigate the presence of *E.coli, K.pneumoniae, S.aureus* bacteria in the membranes of surgeries. The results indicated that E.coli bacteria are the most isolated bacterial species in the current study, if they rank first among bacterial species that take the digestive system as a natural habitat, the resulting yeast may be the cause of internal source contamination such as surgery for the oligmony or appendix, or be external acquired as hospitals, and appeared with K.pneumoniae bacteria isolated from patients with inflammation of surgical operations by (12.34%), It is an opportunistic bacteria because it has many fierce factors that enable it to resist the antibacterial serum, since the natural location of its presence is the upper section of the respiratory system, and as it appeared bacteria *S.aureus* by (9.74%), if it is one of the most important opportunistic bacterial races and its presence in the surgical site does not constitute any shock as it is responsible for most cases of injuries and ulcers. In this study, 59 isolations were tested, divided between (25) *E. coli* (19) ciliary isolation, while *Staphylococcus aureus* was (15) isolation, to determine the susceptibility of bacteria to the production of the membrane by the method of microcalculation dishes, the results showed 30(50.84%) isolation created membrane Strong biosphere, 13 (22.03%) of insulation produced an average biosphere, 14 (23.72%) isolation produced a weak life membrane, the study showed 100% bacteria sensitivity to anti-Ceftriaxone for bacterial species *E.coli, K.pneumoniae, S.aureus,* And 100% Cefazolin for *E.coli, K.pneumoniae.*

1. Introduction

The skin is the first line of defense to defend the body against the invasion of germs because it has fatty acids and factors that help it prevent injury and the growth of organisms that cause diseases, severe and chronic injuries to the skin, leading to a defect in the mechanical functioning of the skin and will allow the opportunity to collect germs that cause inflammation of wounds, The presence of air bacteria K.pneumoniae, E.coli, S.aureus, is considered negative and positive bacterial bacteria for the pigment of Kram and in various forms, if they are conjugal or single or in the form of chains or clusters and classified Depending on the color you turn in the coloring method developed by Hans Christan Gram in 1884 (Sizar&Unakal,2019), one of the most important opportunistic nurses is that it is a widespread polluting factor in hospitals and the acquisition of these bacteria for new qualities enhances their satisfaction (Ubeda et al., 2010), The positive corroids of kram dye of various kinds, including staphylococcus aureus, have the ability to penetrate the defenses of the body and invade its tissues because of its possession factors increase its ferocity and resistance to antibiotics and this was the cause of many diseases of man and these bacteria have become resistant in a way Increased antibiotics, causing a serious global threat (World HealthOrganization,2016), where they have been a major cause of death in hospitals, as the incidence of staphylococcus aureus increases for those who are lying or coming to hospital, as the incidence of S bacteria. Aureus causes infections of wounds and inflammation of caesarean sections, and the formation of pimples, ulcers and inflammation of burns, as well as can cause osteoarthritis, meningitis, blood perch, pneumonia and ulceration of the pleural membrane, as well as the natural fluorescence of the nose and skin of healthy people (Tong et al.,2015), and negative substances of kram dye, including Enteroberiaacte bacteria (Enteroberiacae) , If the bacillus has a portfolio, optional anaerobic living, and grows at a temperature of 37 m, it has the ability to grow in a wide thermal range between (15-45 m), its colonies are dry and pink on the center of Macconkey, to ferment lactose sugar, you can These bacteria produce gas within 48 hours at 35 m temperature (Tenaillon O et al., 2010) and Klebsiella, where it occupies second place and is a member of the intestinal family in terms of medical importance, if it comes after Escherichia coli, which is more common than other nurses causing urinary tract infection and wound inflammation after Gupta et al., 2014, and the bacteria are characterized by the presence of the outer layer, which It consists of an acid capsule containing polysaccharide and is one of the most

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important factors of ferocity responsible for its pathogen (Mawdsley et al., 2019). Biometric membrane is an important factor in enhancing the protection of the disease as it plays a role in the development of brains and reduces the influence of the drug, thereby increasing the resistance of bacteria to antibiotics and converting acute to chronic (Niemirowicz et al., 2014), as the increase in biogastric infection has prevented and removed biometric membranes from the site of bacterial infection to discover strategies such as the formation of biometric membrane in different steps or the use of chemicals (Kostaki et al., 2013; Nostro et al.,2013).

2. Materials and Working Methods

The study was conducted at the hospital between October 4, 2021, and February 1, 2022, when it collected 154 samples of patients with symptoms of surgical inflammation (births). Caesarean sections - tonsils - appendix - colon - hemorrhoids) reviewed and lying at Ramadi Teaching Hospital and Women's and Children's Hospital in Ramadi, the study included the collection of samples of patients who are patients and referred to the hospital by passing and rotating the cotton scanner (Swap cotton), and then transferred the samples To the laboratory and planted on the center of the Maconkey dens and the middle of the blood dens, and in the middle of the blue ideals eosin hugged the dishes antennaally at a temperature (37)°m for (18-24) hours, An hour later, the isolations were subjected to microscopic examination using a cram dye and chemical tests were conducted and all bacterial isolations were diagnosed by VITEK2 Compact. 23%) Esherichia coli and clapsella19 (12.34%) Klebsiella Pneumoniae and Staphylococcus aureus15 (9.74) Staphylococcus aureus in addition to the rest of the isolations that appeared in the study.

3. Results and Discussion

The results of the bacterial implant showed that (124) samples (i.e. 80.52%) gave positive growth in the circles of Maconky akar- Eocin, the Blue Methylation - blood dens), while (30) samples (i.e. 19.48%) did not show growth even after a period of incubation of (48) hours, an If positively developed samples are distributed as follows: caesarean sections(22), hemorrhoid samples (11), appendix (10), colon,9 sample, tonsils(7) sample may be the cause of non-growth in (19.48%) of isolations to inappropriateness The conditions of the air brood, or you may need implants that are not used in the current study, or the cause of infection is virussi, fungal or anaerobic bacteria. The results of the bacterial implant showed that (124) samples (i.e. 80.52%) gave positive growth in the circles of Macconkay akar-Eocin, the blue-like Eocin-blood dens, while (30) samples (i.e. 19.48%) did not show growth even after a period of 48 hours, if the samples were distributed positively Growth as follows: Caesarean sections (22), hemorrhoid sample (11), appendix (10), colon (9), tonsil sample (7), table sample no. 1-1) and form (1-1) may be the cause of non-growth in (19.48%) From isolation to inadequate air hug conditions, or you may need implants unused in the current study, or the cause of infection is virussi, fungal or anaerobic bacteria.

Table 4 Isolated Bacterial Isolations and Their Preparation and Percentage

bacterial isolates	number of isolates	Percentage
E. coli	25	16.23%
Klebseillapneumoniae	19	12.34%
Staphylococcus aureus	15	9.74%
S.epidermidis	11	7.14%
S.lugdunensis	12	7.79%
S.haemolyticus	2	1.30%
S.lentus	3	1.95%
Pseudomonas aeruginosa	12	7.79%
Proteus mirabilis	9	5.84%
Enterobacter	7	4.55%
Acinetobacterbaumannii	9	5.84%
No groth	30	19.48%
	154	100.00%

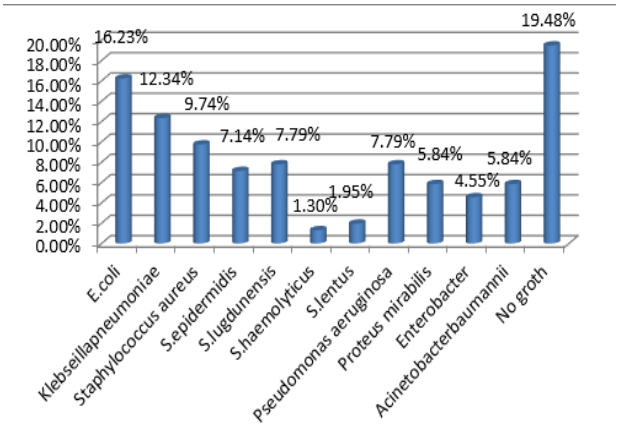

Figure 1 shows isolated bacterial insulation, preparation and percentage

Resistance to E. coli antibiotics

incubated (25) isolation from E.coli bacteria to test allergies to (10) antibiotics, if the results were mixed, if the results were determined by measuring the diameter of the inhibition area (tablet method) and the results were compared according to 2021 /CLSI the results showed a clear difference in the response of bacterial insulation with antibiotics, as shown in the figure (3-6), if E.coli bacteria gave high resistance to the anti-Erythromycin, which belongs to the group of macrolides, if the resistance rate (80%) came This result is consistent with the findings of (pricop et al., 2015 and Fatokum and Onuoha, 2014). It also came close to the conclusion of the mechanism (Al-Dulaimi,2021), if the resistance to isolation (79.2%), while this result did not reconcile with the mechanism (Farsimadan & Kafilzadeh), 2016), if the resistance to isolation (52.8%). While the result of resistance to Tetracycline's E.coli bacteria (72%) as shown in figure 3-6, the reason for the high resistance was due to the possession of efflux pump

bacteria for RNA, in addition to its ability to change the permeability of its outer membrane, as well as the target position switching mechanism, and most E.coli anti-tetracycline insulation is characterized by its ability to produce biometric membranes (Kapoor, 2017) This result was consistent with the conclusion of(tajbakhsh et al., 2016), noting that the rate of resistance to isolations was 75 percent, while gentamicin was (28%), which was consistent with the result of the researcher (Rameiz-Castillo et al., 2018) when it was (28%).

Table 1 Percentage of antibiotic-resistant E. coli isolations						
	Resistance	%	Sensitive	%	Intermediate	%
Ceftriaxone	25	100.00%	0	0.00%	0	0.00%
Cefazolin	25	100.00%	0	0.00%	0	0.00%
Levofloxacin	20	80.00%	5	20.00%	0	0.00%
Erythromycin	20	80.00%	5	24.00%	0	0.00%
Ciprofloxacin	19	76.00%	6	24.00%	0	0.00%
Nitrofurantion	0	0.00%	20	80.00%	5	20.00%
Gentamicin	7	28.00%	18	72.00%	0	0.00%
Cefoxitin	23	92.00%	2	8.00%	0	0.00%
Tetracycline	18	72.00%	7	28.00%	0	0.00%
Vancomycin	19	76.00%	6	24.00%	0	0.00%

K.pneumoniae antibiotic resistance

An allergy (19) bacterial isolation of K.pneumoniae to (10) antibiotics was tested, the results of the current study showed that most of the isolations of the pneumococcal bacteria under study were resistant to antibiotics, the proportion of K.pneumoniae isolations resistant to Tetracycline (89,47) This result was close to that of(Charuak., 2017), and the reason for the high resistance to this antibiotic may be due to the permeability of the outer membrane, or the excessive indiscriminate use of this antibiotic, while the result of the gentamicin allergy test (15.79%), was identical to what(Hu et al., 2016), reached 17.5%, while(Iroha et al., 2009), with resistance (92%), and Erythromycin resistance (68.42%), This is consistent with the findings of the mechanism (Jafer,2017) if the resistance rate (60%), but the reason for the resistance of the isolation of the dogs to this antibiotic may be due to the presence of resistance genes that encrypt the change of location to which the antibiotic is associated, and therefore not The ability of an antibody to affect bacteria (Reygaert,2013), while the resistance to Ciprofloxacin (21.05%) was approximately the mechanism (Hassan,2021) K.pneumoniae showed a high sensitivity to nitrofurantion (68.42%), and the high resistance shown by the dog Sila to Ceftriaxone and by (100%), this result is somewhat similar to the mechanism (Moghadam et al., 2020).

Table 2 shows the percentage of K.pneumoniae antibiotic-resistant and antibiotic-sensitive bacteria						
	Resistance	%	Sensitive	%	Intermediate	%
Ceftriaxone	19	100.00%	0	0.00%	0	0.00%
Cefazolin	19	100.00%	0	0.00%	0	0.00%
Levofloxacin	3	15.79%	16	84.21%	0	0.00%
Erythromycin	13	68.42%	6	31.58%	0	0.00%
Ciprofloxacin	4	21.05%	15	78.95%	0	0.00%
Nitrofurantion	13	68.42%	2	10.53%	4	21.05%
Gentamicin	3	15.79%	14	73.68%	2	10.53%
Cefoxitin	3	15.79%	16	84.21%	0	0.00%
Tetracycline	17	89.47%	2	10.53%	0	0.00%
Vancomycin	17	89.47%	2	10.53%	0	0.00%
Resistance to antibiotic bacteria S.aureus						

The allergy test (15) was conducted with bacterial isolation, which was diagnosed with S.aureus (10) antibiotics, which was the result of the current study of the antibody Ciprofloxacin, which was close to the mechanism (Suod,2005), noting that the resistance rate was 71 As well as the researcher (Al-kazaz,2014), who received a resistance rate (100%) to this antibiotic, but resistance to the bacteria S.aureus for tetracycline, with a resistance rate (69.7%), as noted (Suod, 2005) the resistance rate for this antibiotic was 60 percent, and the reason for the resistance to this antibiotic is the inability of the bacteria S. aureus to assemble the antibody as a result of the presence of (R Factor) As for the antibiotic Ceftriaxone, the resistance rate of 100% was identical to the mechanism (Suod, 2005), while the result of this study for gentamicin antibody is not in line with the findings of the researcher (Matallah et al .,2019) stating that the

resistance rate for this antibiotic was (20,20) 83 percent, while the result of an allergy test for Trimethoprim (26.67%) was similar to what (Al-Khafaji,2018)with a resistance rate of 39.1 percent, and nitrofurantion testing (26.67 percent). It was contrary to his findings (Ragbetli et al.,2016), where the rate of resistance of bacteria to this antibiotic result of poor permeability of its outer membrane or excessive and random use of antibiotics, which encouraged mutations through which the bacteria were resistant, or because of the effect of DNAgyrase or even the loss of proteins associated with penicillin (Fernandez et al., 2010).

Table 3 Percentage of antibiotics for S.aureus bacteria

	Resistance	%	Sensitive	%	Intermediate	%
Ceftriaxone	15	100.00%	0	0.00%	0	0.00%
Cefazolin	4	26.67%	11	73.33%	0	0.00%
Levofloxacin	14	93.33%	1	6.67%	0	0.00%
Trimethoprim	4	26.67%	11	73.33%	0	0.00%
Ciprofloxacin	12	80.00%	3	20.00%	0	0.00%
Nitrofurantion	4	26.67%	11	73.33%	0	0.00%
Gentamicin	8	53.33%	4	26.67%	3	20.00%
Cefoxitin	13	86.67%	2	13.33%	0	0.00%
Tetracycline	12	80.00%	3	20.00%	0	0.00%
Vancomycin	11	73.33%	4	26.67%	0	0.00%

Biofilm production

The results of the study showed a variation in the ability of bacterial insulation under study to produce the biometric membrane, and the results varied in the severity of the composition between weak, medium and strong, that the process of detecting the production of biometric membranes depends on several components including (the type of medium used - the type of surface - the method of detection - conditions of the barn), as the method of microcalcale dishes is characterized by the fact that the surface of the polystyrene used is better in the production of bio-membranes with a stronger effect than the surface of the wire used in the method of catheter tube (Glensk et al., 2019), so I elected this method to investigate the production of the biometric membrane in the current study.

Figure (2) Biofilm production of S. aureus bacteria

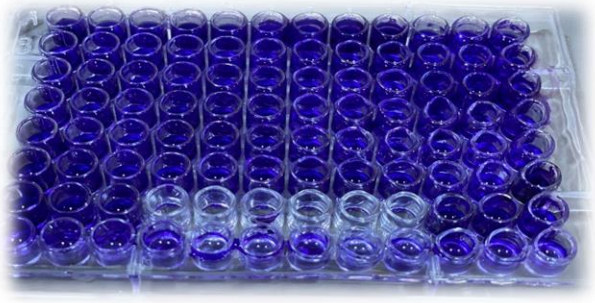

Figure (3) Biofilm production of E. coli bacteria, K.pneumoniae

The results described in Figure 2, (3) and Table (5) and (6) showed that (30) bacterial isolation out of (59) isolation was strongly producing the biosphere, and these isolations differed in the intensity of their absorption when reading their results in the Eliza device, as they were characterized by *E.coli* bacteria are the most bio-membrane-producing bacterial insulation, with 14 of the 25 isolations producing a strong biosphere, while (9) isolations from *K.pneumoniae* bacteria were strongly membrane-producing, and (7) isolations From s.aureus bacteria strong in their production of bio membrane, while biogastric insulation was distributed moderately to (3) isolations from *E.coli* bacteria, (5) isolations from *K.pneumoniae* and (5) bacteria From the bacteria S.aureus, the bacterial insulation that produced the membrane weakly was (8) seclusions of *E.coli* bacteria and (3) isolations from *K.pneumoniae* and (3) from *S.aureus* bacteria.

The variation of bacterial insulation in its ability to produce the biometric membrane may be associated with several factors, including the conditions of the environment surrounding bacterial isolations (temperature- hydrogen number - food availability), as well as a strong association between the ability to produce the biometric membrane and diseases Bacterial insulation (Margues *et al.*, 2017), the method of precision calibration dishes is one of the most accurate and sensitive methods of distinguishing between the amount of production and the study of the early stages of membranes as they use static conditions (fixed), and can be used in Investigate the necessary requirements for the production of biomethics (enzymes - cilia-sodomy) (Saxeno et al., 2014).

Table (5) Production of biometric membrane on MTP microcalculation dishes for *E. coli* bacteria, *K. p S. aureus* wavelength (630nm)					
Isolation numbers for E. coli bacteria	Average optical density (OD) accurate calibration method	*Isolation numbers for S. aureus bacteria*	Average optical density (OD) accurate calibration method	Isolation numbers for K.*pneumoniae bacteria*	Average optical density (OD) accurate calibration method
E. coli 1	0.516	*S. aureus* 1	0.507	*K. pneumoniae* 1	0.576
E. coli 2	0.496	S. aureus 2	0.196	K. pneumoniae 2	0.61
E. coli 3	0.507	S. aureus 3	0.473	K. pneumoniae 3	0.715
E. coli 4	0.614	S. aureus 4	0.222	K. pneumoniae 4	0.603
E. coli 5	0.655	S. aureus 5	0.618	K. pneumoniae 5	0.381
E. coli 6	0.212	S. aureus 6	0.215	K. pneumoniae 6	0.573
E. coli 7	0.665	S. aureus 7	0.778	K. pneumoniae 7	0.396
E. coli 8	0.311	S. aureus 8	0.411	K. pneumoniae 8	0.262
E. coli 9	0.196	S. aureus 9	0.575	K. pneumoniae 9	0.722
E. coli 10	0.354	S. aureus 10	0.446	K. pneumoniae 10	0.368
E. coli 11	0.692	S. aureus 11	0.600	K. pneumoniae 11	0.292
E. coli 12	0.569	S. aureus 12	0.369	K. pneumoniae 12	0.633
E. coli 13	0.560	S. aureus 13	0.507	K. pneumoniae 13	0.477
E. coli 14	0.614	S. aureus 14	0.350	K. pneumoniae 14	0.710
E. coli 15	0.424	S. aureus 15	0.602	K. pneumoniae 15	0.444
E. coli 16	0.223			K. pneumoniae 16	0.535
E. coli 17	0.569			K. pneumoniae 17	0.253
E. coli 18	0.293			K. pneumoniae 18	0.0741
E. coli 19	0.556			K. pneumoniae 19	0.092
E. coli 20	0.185				
E. coli 21	0.505				
E. coli 22	0.311				
E. coli 23	0.582				
E. coli 24	0.210				
E. coli 25	0.788				

Table 6 Percentage of biometric production of E.coli bacteria, K.p S.aureus				
Percentage (%)				Type of bacteria
The method of precision calibration dishes MTP				
unproductive	Average	weak	Strong	
	12.00%	32.00%	56.00%	E. coli
10.53%	26.32%	15.79%	47.37%	k.p
	33.33%	20.00%	46.67%	S.aureus

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