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Isolation, identification and antibiogram studies on *P. aeruginosa* and *S. aureus* from wound samples in and around Tirupati

PV Siddartha, V Kalpana, A Jagadeesh Babu, CS Swetha and T Madhava Rao

Abstract
The present study was carried out to detect the pyogenic microorganisms from the wounds of animal origin, the antimicrobial resistant patters in and around Tirupati region. This study addresses prevalence of 2 different organisms *i.e., S. aureus* and *P. aeruginosa* by cultural and biochemical characterization from pus and wound infections and their antimicrobial resistance to different antibiotics commonly used against them. A total of 150 samples of wound and skin affections were collected from various animal species including cattle, buffalo, goats and sheep, irrespective of gender and age. Among them, 71 *Staphylococcus* isolates were identified and subjected to antibiotic sensitivity test using 9 different and most commonly used antibiotics. The isolates have shown maximum sensitivity towards gentamicin (83.09%) followed by co-trimaxazole (74.65%) and least sensitive to erythromycin (14.08%). Among the isolates of *Staphylococcus aureus* highest resistance was observed to penicillin G (78.87%) followed by erythromycin (63.38%) and least resistant to Gentamicin (9.86%). Among 150 samples, 18 samples were identified as *Pseudomonas* isolates and were subjected to antibiotic sensitivity test. The isolates have shown maximum sensitivity to chloramphenicol and amikacin (66.67%) and least to erythromycin (11.11%). Further these isolates have shown intermittent resistance to gentamicin (22.22%) and amikacin (11.11%). The *P. aeruginosa* isolates have shown maximum resistance to erythromycin (88.88%) and least resistance to gentamicin and amikacin (22.22%).

Keywords: *S. aureus*, P. aeruginosa, pyogenic microorganisms, ABST

Introduction
A wound is a type of injury which happens relatively quickly in which skin is torn, cut or punctured (an open wound), or where blunt force trauma causes a contusion (a closed wound). Most wounds heal without complications, however, the care of wounds in animal species can be a challenging endeavour. The presence of debris, dirt, hair, suture and necrotic or devitalized tissue act as foreign material, leading to an intense inflammatory reaction that prolongs the inflammatory phase and delays the repair phase. Accumulation of fluid in the wound be encourages infection and leads to wound ischemia, delaying wound healing. Exposure of the underlying tissue following a loss of skin integrity due to a range of reasons provides a moist, warm, and nutritious environment that is favourable to microbial colonization and proliferation. Wound infection is one of the health problems that are caused by various types of pathogens. Wound colonization is most frequently poly-microbial, involving different microorganisms that could be potentially pathogenic.

Pus is a whitish yellow material made primarily of dead white blood cells and dead Bacteria which is normally found in regions of bacterial infections produced during inflammatory pyogenic bacterial infections. There are different microbial species which are responsible for the pus formation in wounds, ear infections and brain abscess. These include different bacterial and fungal species. The most common pus producing bacteria are *Staphylococcus aureus (S. aureus), Klebsiella spp., Pseudomonas, Escherichia coli, streptococci*. *S. aureus* is most common bacteria that produce pus (Singh, 2014) [21]. *Pseudomonas* infections also constitute a major health care problem because this pathogen has developed resistance to most antibiotics introduced in antibiotherapy and is also one of the most predominant bacteria in pus producing wounds. The human skin and soft tissue infections that are caused by microbes in many cases result in the production of pus, a whitish yellow fluid comprised of dead WBCs, cellular debris, and necrotic tissues (Cogen, 2008; Scalise, 2015) [7, 20].

In this study, the *P. aeruginosa* and *S. aureus* have been isolated, characterized and their Antibiogram profile was studied from wound samples that have been collected from Tirupati region.

Materials and Methods

The present study was carried out in the Department of Veterinary Public Health and Epidemiology, College of Veterinary Science, Tirupati.

Sterilization of glassware

The glassware used in this study was made by Borosil India Ltd, Mumbai. Sterilization of glassware was done by soaking them in a Labolene solution overnight. Then, the glassware was washed thoroughly in running tap water, followed by soaking them in an acidic solution for 2 hrs. Then the glassware was rinsed three times with tap water and then immersed in triple-distilled water overnight. The air-dried glassware was packed and sterilized in hot air oven for one hour at 160 ^{0}C.

Sterilization of plastic ware

Plastic ware and rubber items were sterilized by autoclaving at 121^{0}C at 15 psi pressure for 15 minutes.

Collection of samples

A total of 150 samples of wound and skin affections which were collected from various animal species including cattle, buffalo, goats and sheep, irrespective of gender and age. Pus swabs were also obtained from wound sites. The specimens were collected on sterile cotton swab without contaminating them with skin commensals. Each specimen was collected by rotating a sterile, pre-moistened swab across the wound surface of a 1 cm^2 area in a zig-zag motion, from the centre to the outside of the wound. Then, the swab was placed in the tube containing the sterile normal saline The swab samples were directly brought to the Department of Veterinary Public Health and Epidemiology, College of Veterinary Science, Tirupati for further laboratory processing by standard procedures. In the laboratory, the specimens were registered and swabs were cultured on nutrient broth and incubated at 37ºC for 24 h.

Isolation of organisms

Collected swab samples were processed for isolation and identification of different pathogenic bacteria according to the standard operating procedures of the laboratory. Upon receipt, as per the history of the sample each swab was inoculated into nutrient broth (NB) and accordingly incubated at 370C up to 24-48 hrs. before attempting pure colony isolation over solid nutrient medium. After incubation, A loop full of inoculum was streaked on Mannitol Salt Agar and Cetrimide agar for *Staphylococcus aureus* and *Pseudomonas aeruginosa* respectively. The plates were then incubated at 37^{0}C for 18-24 hrs. After incubation, the organism specific colonies were picked up and inoculated on nutrient agar slants and further confirmation was done by biochemical characterization of the isolates as per the methods described by Cruickshank, (Medical Microbiology, Vol. II, 12th Ed, 1975) [8].

Bacterial smears were prepared by mixing typical colonies with sterile saline on a clean glass microscopic slide. The smear was allowed to air dry and heat-fixed over the flame. The slide was then stained with Grams method of staining and examined under 100X oil immersion lens of the microscope. The organisms were identified based on their morphological characteristics.

Biochemical characterization

The biochemical tests conducted for confirmation of *S. aureus* and *P. aeruginosa* were catalase test, oxidase test, urease test, IMViC tests, and the nitrate reduction test. These tests were conducted as per the methods described by Cruickshank, (Medical Microbiology, Vol. II, 12th Ed, 1975) [8].

In-vitro antibiotic sensitivity test

Bacterial suspension was made by transferring 4-5colonies from primary isolated medium i.e Mannitol Salt agar and Cetrinide agar to 5 ml of nutrient broth by touching the top of the colonies with a flame sterilized and cooled platinum loop. The resulting culture after incubation at 37^{0}C for 8 hours was compared with the turbidity standard prepared separately for adjustment of bacterial suspension. Commercially available standard antimicrobial discs were procured and stored at 2-8^{0}C in the refrigerator. Unopened disc containers were removed from the refrigerator 1-2 hours before use, to bring them to room temperature. The anti-microbial dics with known concentrations as noted on micrograms(µg) or International Units (IU) per disc were used to study the antimicrobial susceptibility of the isolates.

Recovered bacterial isolates were subjected to antibiotic sensitivity testing against 9 different antimicrobial discs (Table 1) to assess the patterns of antibiotic activity against the bacteria isolated from the wound samples.

Table 1: List of antibiotics used for the antibiotic susceptibility test

S. No.	Name of Antibiotic disc	Symbol	Concentration
1.	Ceftriaxone	CTR	30µg
2.	Chloramphenicol	C	30µg
3.	Tetracycline	TE	30µg
4.	Erythromycin	E	15µg
5.	Gentamicin	GEN	10µg
6.	Co- Trimaxazole	COT	25µg
7.	Ciprofloxacin	CIP	5µg
8.	Penicillin	P	10 µg
9.	Amoxicillin	AMX	10 µg

Results

Isolation of *Staphylococcus*

All the wound samples collected from different animals were inoculated into nutrient broth and incubated at 37^{0}C for 24 hrs. After the incubation period a loop full of inoculum from nutrient broth tubes was streaked on Mannitol salt agar plates by following the aseptic precautions. The plates were incubated at 37^{0}C for 48 hrs. Then the plates were observed for Yellow colour colonies with yellow zones on Mannitol salt agar plates. (Fig 1). Among the 150 samples 75 samples were positive for Staphylococcus by culture method and were subjected to grams staining and found Gram positive, cocci shaped organisms arranged in clusters (bunch of grapes). (Fig 2).

The isolates were subjected to the biochemical tests like catalase test, oxidase test, urease test, nitrate reduction test, indole test, methyl red test, vogues proskauer test and citrate utilization test. The observations in the biochemical tests are as in the (Table 2, Fig 3-7).

Table 2: Results of the confirmatory biochemical tests for Staphylococcus

Tests	No. of samples	Positive by cultural method as *Staphylococcus*	No. of isolates confirmed as *S.aureus* after biochemical tests	Result
Samples analysed	150	75	71	71
Biochemical tests				
Test	**Original colour of the medium**	**Positive reaction**	**Identification index of *S.aureus***	**Result**
Catalase test	Colourless	Appearance of bubbles	Appearance of bubbles	Positive
Oxidase test	Colourless	Blue	Negative	Negative
Indole test	Colourless	Red ring	Negative	Negative
Methyl red test	Colourless	Red	Positive	Positive
Voges Proskauer test	Colourless	Deep rose colour	Positive	Positive
Citrate utilization test	Green	Prussian blue	Positive	Positive
Nitrate reduction test	Colourless	Red	Positive	Positive
Urease test	Straw colour	Pink	Positive	Positive

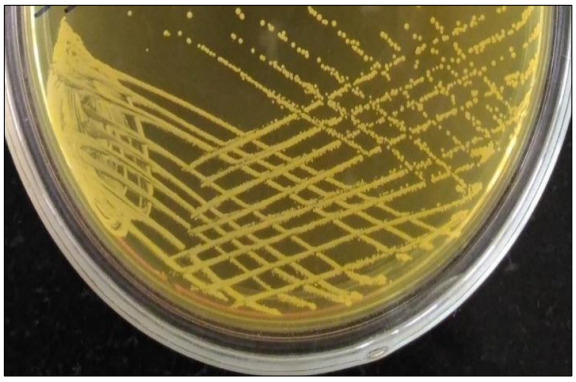


Fig 1: *Staphylococcus aureus* showing golden yellow colour colonies

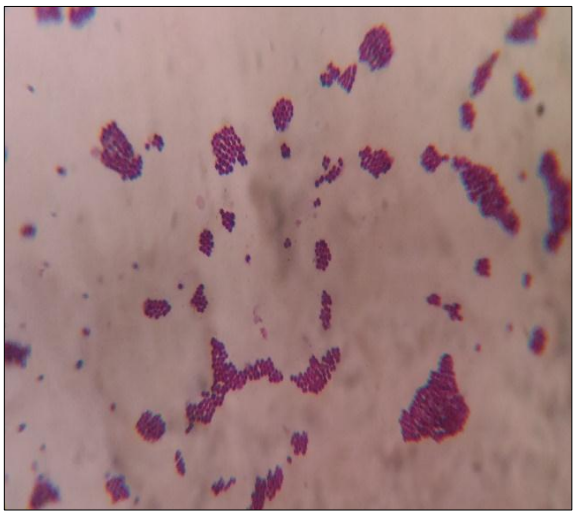


Fig 2: Grams staining showing characteristic gram positive coccci of *S. aureus*

Fig 3: Catalase positive reaction – *Staphylococcus* spp

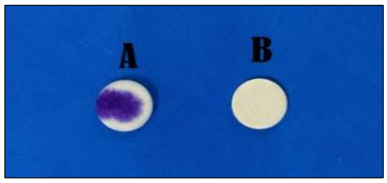


Fig 4: *Staphylococcus spp.* (Oxidase negative) A-Oxidase positive B-Oxidase negative

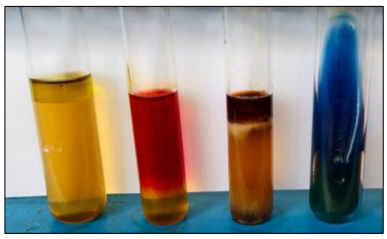


Fig 5: IMViC Reactions (-, +, +, +) for *S. aureus*

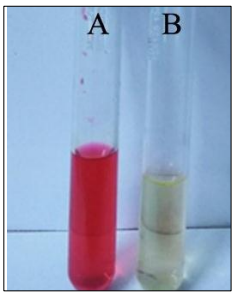


Fig 6: Tube A- Nitrate positive reaction (*Staphylococcus* spp.) Tube B- Nitrate negative reaction

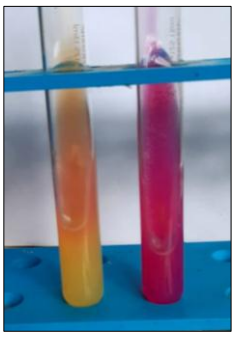


Fig 7: Tube-A Urease negative tube-B Urease positive (*Staphylococcus* spp.)

Isolation of *Pseudomonas*

All the wound samples collected from different animals were inoculated into nutrient broth and incubated at 37^0C for 24 hrs. After the incubation period a loop full of inoculum from nutrient broth tubes was streaked on Cetrimide agar plates by following all the aseptic precautions. The plates were incubated at 37^0C for 48 hrs. Then the plates were observed for Greenish blue colonies characterized by irregular growth on Cetrimide agar plates (Fig 8). Among the 150 samples 21 samples were positive for Pseudomonas by culture method and were subjected to grams staining and found Gram negative, rods either straight or slightly curves. (Fig 9)

The isolates were subjected to the biochemical tests like catalase test, oxidase test, urease test, nitrate reduction test, indole test, methyl red test, vogues proskauer test and citrate utilization test. The observations in the biochemical tests are as in the (Table 3 and Fig 10-12).

Table 3: Results of the confirmatory biochemical tests for *pseudomonas*

Tests	No. of samples	Positive by cultural method as Pseudomonas	No. of isolates confirmed as Pseudomonas after biochemical tests	Result
Samples analysed	150	21	18	18
Biochemical tests				
Test	**Original colour of the medium**	**Positive reaction**	**Identification index of pseudomonas**	**Result**
Catalase test	Colourless	Appearance of bubbles	Appearance of bubbles	positive
Oxidase test	Colourless	blue	positive	positive
Indole test	Colourless	Red ring	Negative	Negative
Methyl red test	Colourless	Red	Negative	Negative
Voges Proskauer test	Colourless	Deep rose colour	Negative	Negative
Citrate utilization test	Green	Prussian blue	Positive	Positive
Nitrate reduction test	Colourless	Red	Positive	Positive
Urease test	Straw colour	pink	Negative	Negative

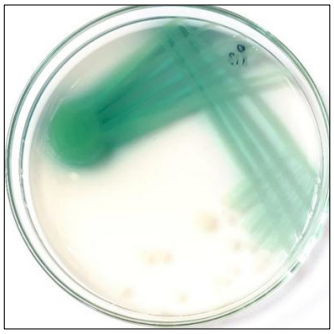


Fig 8: Greenish blue colonies on cetrimide agar

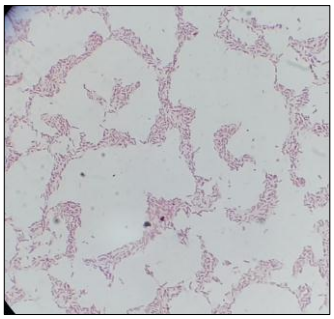


Fig 9: Grams staining showing characteristic gram negative rods of *Pseudomonas aeuriginosa*

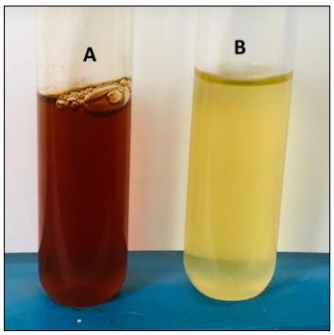


Fig 10: Tube-A (Positive nitrtate reduction in *Pseudomonas aeruginosa)*

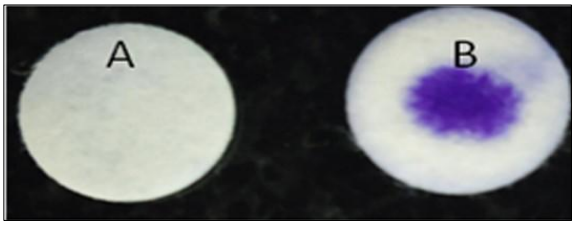


Fig 11: *Pseudomonas aeruginosa* showing positive oxidase reaction Disc A Negativecontrol Disc-B positive reaction

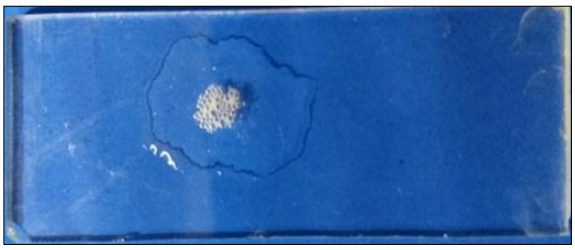


Fig 12: *Pseudomonas aeruginosa* showing positive catalase reaction

Antimicrobial sensitivity / resistance patterns of the isolates

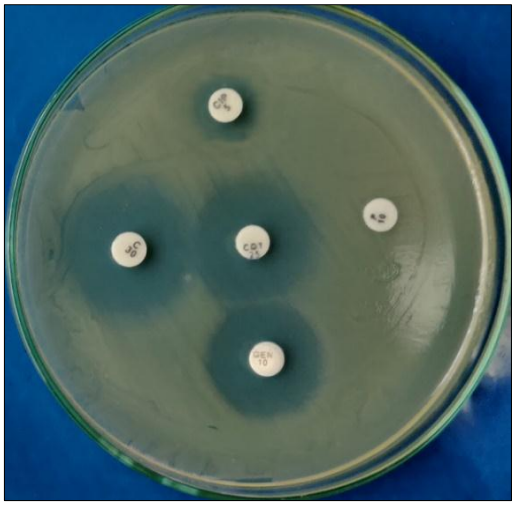
The pathogenic microorganisms which were isolated from the wounds were subjected to antibiotic sensitivity test by using 9 commercially available antibiotic discs by using disc diffusion method.

All the 71 *Staphylococcus* isolates were subjected to antibiotic sensitivity test using 9 different and most commonly used antibiotics in veterinary medicine as mentioned in table. The isolates have shown maximum sensitivity towards gentamicin (83.09%) followed by co-trimaxazole (74.65%), chloramphenicol (73.24%), amoxicillin (53.32%), tetracycline (49.30%), ciprofloxacin (42.25%), ceftriaxone (29.58%), penicillin (21.13%) and erythromycin (14.08%). Notable percentage of isolates have shown intermediate resistance to erythromycin (22.54%) followed by ciprofloxacin, tetracycline (21.13%), ceftriaxone (16.9%), chloramphenicol (15.49), co-trimaxazole (14.08%) and gentamicin (7.04%). Among the isolates of *Staphylococcus aureus* highest

resistance was observed to penicillin G (78.87%) followed by erythromycin (63.38%), ceftriaxone (53.52%), amoxicillin (46.48%), ciprofloxacin (36.62%), tetracycline (29.58%), co-trimaxazole (11.26%), chloramphenicol (11.26%) and gentamicin (9.86%) as shown in Table 4 and Fig 13-15.

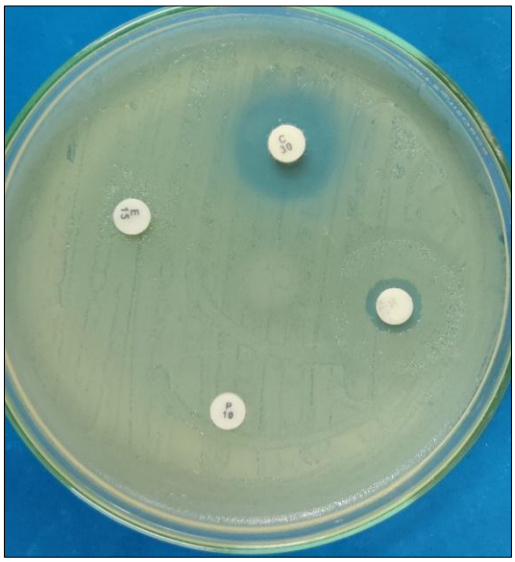
Table 4: Antibiotic sensitivity /resistance patterns of *Staphylococcus*

	Antibiotic disc	No. of isolates		
		Sensitive	Intermediate	Resistant
1.	Ceftriaxone (CTR 30 mcg)	21(29.58%)	12 (16.9%)	38 (53.52%)
2	Ciprofloxacin(Cip 5mcg)	30 (42.25%)	15 (21.13%)	26 (36.62%)
3	Chloramphenicol(C 30mcg)	52 (73.24%)	11 (15.49%)	08 (11.26%)
4.	Gentamicin (Gen 10)	59 (83.09%)	5 (7.04%)	7 (9.86%)
5.	Amoxicillin (AMX 10)	38 (53.32%)	0	33 (46.48%)
6.	Co-trimaxazole (COT-25)	53 (74.65%)	10 (14.08%)	8 (11.26%)
7.	Tetracycline (TE-30)	35 (49.30%)	15 (21.13%)	21 (29.58%)
8.	Erythromycin(E-15)	10 (14.08%)	16 (22.54%)	45 (63.38%)
9.	Penicillin (P 10)	15(21.13%)	0	56(78.87%)



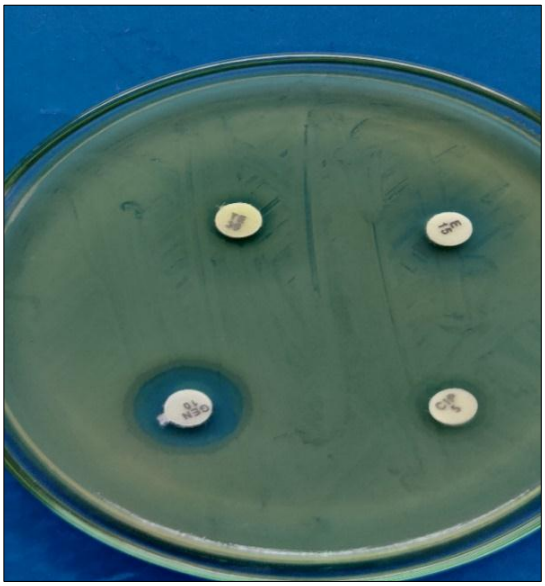
Tetracycline Sensitive
Gentamicin Sensitive
Co-Trimaxazole Sensitive
Ciprofloxacin Sensitive

Fig 13: Antibiotic sensitive patterns of Staphylococci



Gentamicin Intermediate sensitive
Chloramphenicol Intermediate sensitive
Ciprofloxacin Intermediate sensitive

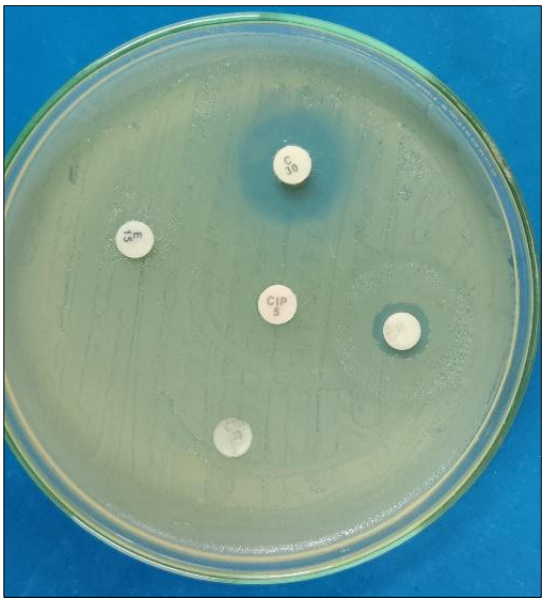
Fig 14: Antibiotic intermediate sensitive patterns of Staphylococci



Penicillin Resistant
Amoxicillin Resistant
Ceftriaxone Resistant

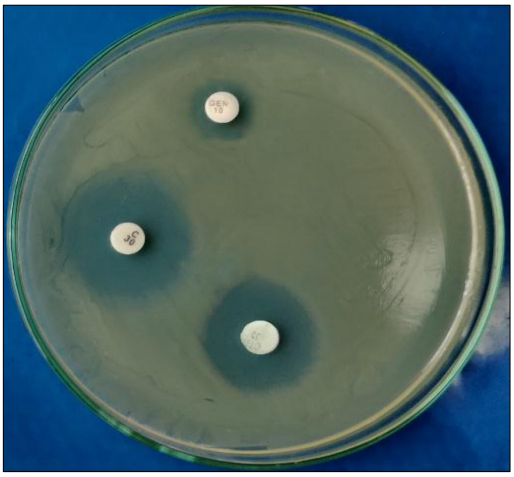
Fig 15: Antibiotic resistant patterns of Staphylococci

All the 18 *Pseudomonas* isolates were subjected to antibiotic sensitivity test using 9 different and most commonly used antibiotics in veterinary medicine as mentioned in table (write from MM). The isolates have shown maximum sensitivity to chloramphenicol and amikacin (66.67%) followed by gentamicin and tetracycline (55.56%), ciprofloxacin (44.44%), Co-trimaxazole (33.33%), ceftriaxone, amoxicillin (22.22%) and erythromycin (11.11%). Further these isolates have shown intermittent resistance to gentamicin (22.22%) and amikacin (11.11%). The Pseudomonas aeruginosa isolates have shown maximum resistance to erythromycin (88.88%) followed by ceftriaxone and amoxicillin (77.78%), co-trimaxazole (66.67%), ciprofloxacin (55.56%), Tetracycline (44.44%) chloramphenicol (33.33%), gentamicin and amikacin (22.22%) as shown in Table 5 and Fig 16-18.



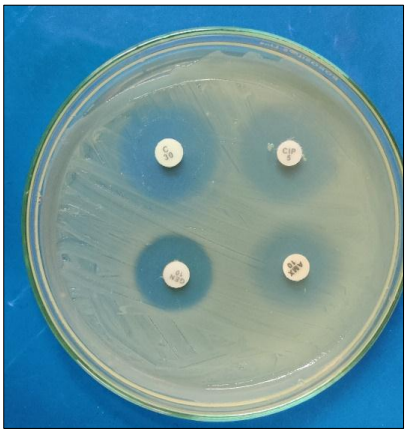
Ciprofloxacin Resistant
Erythromycin Resistant
Ceftriaxone Resistant

Fig 16: Antibiotic resistance patterns of P. aeruginosa



Amikacin Intermediate sensitive
Gentamicin Intermediate sensitive

Fig 17: Antibiotic Intermediate sensitive patterns of P. aeruginosa



Chloramphenicol Sensitive
Gentamicin Sensitive
Amikacin Sensitive

Fig 18: Antibiotic sensitive patterns of *P. aureginosa*

Table 5: Antibiotic sensitivity /resistance patterns of *Pseudomonas*

S. No.	Antibiotic disc	No. of isolates		
		Sensitive	Intermediate	Resistant
1.	Ceftriaxone (CTR 30 mcg)	4 (22.22%)	- (0%)	14(77.78%)
2	Ciprofloxacin(cip 5mcg)	8 (44.44%)	0	10 (55.56%)
3	Chloramphenicol(C 30mcg)	12 (66.67%)	0	6 (33.33%)
4.	Gentamicin (Gen 10)	10 (55.56%)	4 (22.22%)	4 (22.22%)
5.	Amoxicillin (AMX 10)	4 (22.22%)	0	14(77.78%)
6.	Co-trimaxazole (COT-25)	6 (33.33%)	0	12 (66.67%)
7.	Tetracycline (TE-30)	10 (55.56%)	0	8 (44.44%)
8.	Erythromycin(E-15)	2 (11.11%)	0	16 (88.88%)
9.	Amikacin (AMK 10)	12 (66.67%)	2 (11.11%)	4 (22.22%)

Discussion

The findings of the present study concluded that the rapid emergence of resistant bacterial isolates from wounds of animals accentuates the potential impact of antimicrobial resistant organisms. Skin wounds are a common presentation in animal practice. The successful management of wound healing in animals requires knowledge of the physiology of the wound healing process and the application of an appropriate therapeutic intervention. Skin ailments or wounds are probably the most common causes of enhanced susceptibility of animals to infections, as they are prone to bacterial contamination. Occasionally may spread over the body surfaces and may eventually result into internal abscesses, fistula and sometimes septicemia. Skin affections including wound infections are frequent complications following lacerations, injuries, penetrating trauma, animal fights, bites etc. In conventional animal rearing practices, wounds were mostly left ignored to be self-cured until they affect the general condition and productivity of the animals including meat, leather quality, or economy of the owner. If treated without confirmatory diagnosis, untargeted therapy and injudicious use of allopathic drugs, gives way to emergence of antimicrobial drug resistant pathogens (Tiwari *et al.*, 2015) [22].

Many wounds are colonized by bacteria or show signs of clinical infection (Kozar *et al.*, 2018) [14]. The successful management of bacteria in a wound is of great importance; however, it is still a complex issue (Bessa *et al.*, 2015) [5]. Therefore, our study evaluates the current situation in a particular geographic area, which is mostly helpful to the clinicians and microbiologists involved because it can make them aware of the real circumstances that they are dealing with presently. Pyogenic or pus forming wound infections are characterized by severe local inflammation subsequent to tissue injury leading to generalized clinical disease through the various toxic mechanisms associated with invasion of pyogenic bacteria. The crisis of antibiotic resistance among pyogenic bacterial infections has been attributed to the inappropriate use of antimicrobial agents particularly in developing country. The antimicrobial resistance has become a global challenge and the resistant pathogen poses a grave threat to the public health worldwide. Different studies are being conducted across the globe to access the bacterial profile in pyogenic wound infection. The appropriate knowledge of the pathogens, their resistant character, and their updated antimicrobial therapy plays a crucial role in the treatment process as well as in infection control measures. Therefore, this study was intended to characterize the bacterial isolates from clinical specimens of pyogenic wound infections and to determine the antibiotic susceptibilities to commonly used therapeutic regimens. Polymicrobial pyogenic wound infections might be associated with poor wound care, increased microbial survival, and ineffective antimicrobial treatment (Mama *et al.*, 2014) [16]. Trojan *et al.* 2016 [23] have documented the Gram negative bacterial dominance in pyogenic wound infections. On the other hand, *Pseudomonas aeruginosa* (49.2%) was the predominant isolate responsible for pyogenic wound infections in this study which is quite similar to several previous studies (Muluye *et al.*, 2014). It is well known that *S. aureus* and Gram negative bacterial pathogens produce very potent virulence factors, responsible for maintaining the infection and delaying the process of wound healing (Bessa *et al.*, 2015) [5].

Out of 150 pus samples processed the prevalence of S*taphylococcus* was 47.33% which is in accordance with the findings of Damen *et al.* (2015) [9] who have reported 45.2% and Biradar *et al.* (2016) [6] who have found 41.98%. Kumar *et al.* (2020) [15], Agnihotri *et al.* (2004) [1] and Trojan *et al.* (2016) [23] have reported a low prevalence of 15.4%, 17.9% and 21% respectively. Among 150 pus samples processed the *Pseudomonas* was isolated with a prevalence of 12% which is in accordance with the findings of who have reported 12.5% Altoparlak *et al.* (2004) [2] and Guptha *et al.* (2015) 13.1%. The findings were less than Agnihotri *et al.* (2004) [1] who have reported 59%, Duggal *et al.* (2015) [10] reported (29.73%) and Bessa *et al.* (2015) [5] 27% of *Pseudomonas* in the samples and these findings were higher than the present study. The findings of the by Kozer *et al.* (2013), Ayub (2015) [4] and Trojen *et al.* (2016) who have reported 3.33%, 5.8% and 6.58% respectively.

In the present study, Staphylococcus has shown resistance towards amoxicillin was 46.48% and for Erythromycin it was 63.38%. Similar results towards amoxicillin (33.1%) and Erythromycin (72.7%) was reported by Javed *et al.* (2011). In the present study, sensitivity towards Tetracycline was 49.30%. Similar results of 40% sensitivity was observed towards tetracycline by Rao *et al.* (2014) [19]. In contrast, Goswami *et al.* (2011) [11] reported sensitivity of 25% towards tetracycline. In the present study resistance towards amoxicillin and Co-trimaxizole was 46.48% and 11.26% respectively. In contrast, Ohalete *et al.* (2012) [18] reported 97.7% resistance towards amoxicillin and 100% resistance towards co-trimaxozole. In the present study, resistance towards erythromycin was 63.38%. Similar results were reported by Ohalete *et al.*, (2012) [18] in which the resistance was 68.2%. In present study, sensitivity towards gentamicin was 83.09% and 73.24% was for chloramphenicol. In agreement with the results obtained, Biradar *et al.* (2016) [6] reported sensitivity towards gentamicin and chloramphenicol was 89.47% and 68.42% respectively.

In this study, Pseudomonas showed highest sensitivity to Amikacin (66.67%) which was similar to the results of Anjum and Mir (2010) [3] who reported sensitivity of amikacin (79%). In the present study, resistance and sensitivity to Ceftriaxone was 77.78% and 22.22% respectively which was in agreement with the results of Duggal *et al.* (2015) [10] reported the resistance was 72.73% and Tiwari *et al.* (2015) [22] in where 25.64% of sensitivity was observed. Pseudomonas was shown 22.22% sensitivity to Ceftriaxone, which was almost similar with the results of Tiwari *et al.* (2015) [22] who reported 25.64% sensitivity. In the present study, 55.56% sensitivity towards gentamicin was observed. Similar results were reported by Ohalete *et al.*, (2012) [18] in which the sensitivity towards gentamicin was 48.7%. In the present study, sensitivity towards ceftriaxone (22.22%) and ciprofloxacin (44.44%) was observed. Similar results were reported by Biradar *et al.* (2016) [6] in which sensitivity towards ceftriaxone was 16.66% and for ciprofloxacin it was 37.5%. Our findings indicate the existence of high drug resistant bacteria in pyogenic wound infections. The high use of β-lactam antibiotics and cephalosporins and inappropriate infection control procedures in the hospitals might be the cause of rising rates of resistance among these bacteria. Moreover, longer duration of prophylactic antimicrobial exposure may contribute to organisms for developing resistance.

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Conflict of interest

The authors of this paper declares that there is no conflict of interest.

References

1. Agnihotri N, Gupta V, Joshi RM. Aerobic bacterial isolates from burn wound infections and their antibiograms - A five-year study. Burns. 2004;30(3):241-243.
2. Altoparlak U, Aktas F, Celebi D, Ozkurt Z, Akcay MN. Prevalence of metallo-β-lactamase among Pseudomonas aeruginosa and Acinetobacter baumannii isolated from burn wounds and *in vitro* activities of antibiotic combinations against these isolates. Burns. 2005;31(6):707-710.
3. Anjum F, Mir A. Susceptibility pattern of pseudomonas aeruginosa against various antibiotics. African Journal of Microbiology Research. 2010;4(10):1005-1012.
4. Ayub M. Isolation of Pathogens Causing Sepsis, Pus and Infected Wounds from Critical Care Unit: A Retrospective Study. Annals of Clinical and Laboratory Research. 2015;3(4):1-7.
5. Bessa LJ, Fazii P, Di Giulio M, Cellini L. Bacterial isolates from infected wounds and their antibiotic susceptibility pattern: some remarks about wound infection. International wound journal. 2015;12(1):47-52.
6. Biradar A, Farooqui F, Prakash R, Khaqri SY, Itagi I. Aerobic bacteriological profile with antibiogram of pus isolates. Indian Journal of Microbiology Research. 2016;3(3):245.
7. Cogen AL, Nizet V, Gallo RL. Skin microbiota: a source of disease or defence? British Journal of Dermatology. 2008;158(3):442-455.
8. Cruickshank R, Duguid JP, Marmion BP, Swain RHA. Medical microbiology. 12th. Ed. Churchill Livingstone Edinburgh. London, England, 1975.
9. Damen JG, Faruk S, Dancha C. Aerobic Bacteria Isolates of Septic Wound Infections and their Antibiogram in North Central Nigeria. American Journal of Biomedical and Life Sciences. 2015;3(3):36.
10. Duggal S, Khatri PK, Parihar RS, Arora R. Antibiogram of Various Bacterial Isolates from Pus Samples in a Tertiary Care Centre in Rajasthan. International Journal of Science and Research (IJSR). 2015;4(5):1580-1584.
11. Goswami NN, Trivedi HR, Goswami APP, Patel TK, Tripathi CB. Antibiotic sensitivity profile of bacterial pathogens in postoperative wound infections at a tertiary care hospital in Gujarat, India. Journal of pharmacology & pharmacotherapeutics. 2011;2(3):158.
12. Gupta AK, Batra P, Mathur P, Karoung A, Thanbuana BT, Thomas S, *et al.* Microbial epidemiology and antimicrobial susceptibility profile of wound infections in out-patients at a level 1 trauma centre. Journal of Patient Safety and Infection Control. 2015;3(3):126-129.
13. Javeed I, Hafeez R, Anwar M. Antibiotic susceptibility pattern of bacterial isolates from patients admitted to a tertiary care hospital in Lahore. Biomedica. 2011;27:19-23.

14. Kozar M, Hamilton H, Koscova J. Types of Wounds and the Prevalence of Bacterial Contamination of Wounds in the Clinical Practice of Small Animals. Folia Veterinaria. 2018;62(4):39-47.
15. Kumar GS, Sreedevi S, Krishna JB. One year study of aerobic bacterial profile and antimicrobial susceptibility pattern of pus samples. Indian Journal of Microbiology Research. 2020;7(1):1-5
16. Mama M, Abdissa A, Sewunet T. Antimicrobial susceptibility pattern of bacterial isolates from wound infection and their sensitivity to alternative topical agents at Jimma University Specialized Hospital, South-West Ethiopia. Annals of Clinical Microbiology and Antimicrobials, 2014, 13(1).
17. Muluye D, Wondimeneh Y, Ferede G, Nega T, Adane K, Biadgo B, *et al*. Bacterial isolates and their antibiotic susceptibility patterns among patients with pus and/or wound discharge at Gondar university hospital. BMC Research Notes. 2014;7(1):4-8.
18. Ohalete CN, Obi RK, EmeaKoroha MC. Bacteriology of different wound infection and their antimicrobial susceptibility patterns in Imo state Nigeria. World Journal Pharmacy and Pharmaceutical Sciences. 2012;1(3):1155-1172.
19. Rao DR, Basu R, Biswas DR. Aerobic Bacterial Profile and Antimicrobial Susceptibility Pattern of Pus Isolates in a South Indian Tertiary Care Hospital. Surgery. 2014;36:35-2
20. Scalise A, Bianchi A, Tartaglione C, Bolletta E, Pierangeli M, Torresetti M, *et al*. Microenvironment and microbiology of skin wounds: the role of bacterial biofilms and related factors. In Seminars in vascular surgery. WB Saunders. 2015 Sept;28(3-4):151-159.
21. Singh A, Sao S, Aswani J. Isolation Of staphylococcus aureus from wound/swab and its antibiogram study.
22. Tiwari R, Yadav SK, Singh S, Gangwar NK. Bacterial etiology of skin and wound infections along with antibiogram profiling in reference to emerging antibiotic resistance in animals. Advances in Animal and Veterinary Sciences. 2015;3(5):259-268.
23. Trojan R, Razdan L, Singh N. Antibiotic susceptibility patterns of bacterial isolates from pus samples in a tertiary care hospital of Punjab, India. International journal of microbiology, 2016.