



Development of Antibigram for Secondary Health care Hospital

Anand Vijayakumar PR^{*1}, Lalramengmawii², Lalduhawmi TC², Manisha S², Shekhar S Deshpande²

¹Department of Pharmacology, JSS College of Pharmacy (JSS Academy of Higher Education & Research, Mysuru), Ooty-643001, Tamil Nadu, India

²Department of Pharmacy Practice, JSS College of Pharmacy (JSS Academy of Higher Education & Research, Mysuru), Ooty-643001, Tamil Nadu, India



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ABSTRACT

Antibiotics resistance is an emerging problem in the management for infectious diseases. Patients are many a time prescribed with antibiotics without knowing that particular antibiotic sensitivity pattern with respect to the infectious microorganism. This study aims to detect the type of microbes causing certain infections in the hospital and also to detect the sensitivity pattern of the antibiotics to these microbes. We conducted a prospective study for six months on the neonates who were admitted in NICU. The blood samples were collected from these neonates before the administration of antibiotics. The swab samples were also collected from various places of this hospital to detect the types of microorganisms present in the hospital and to study the sensitivity of the antibiotics toward these microbes. The antibiotics used in this study were Gentamicin, Ampicillin, Cefotaxime, Amikacin, Piperacillin, Meropenam, and Vancomycin. *Staphylococcus aureus* and *Streptococcus pneumoniae* were found to be the most common pathogens implicated in neonate's infection. All the organisms showed absolute sensitivity mostly to Ampicillin, Gentamicin, and Piperacillin and resistant to Cefotaxime, Amikacin, and Vancomycin. *Staphylococcus aureus*, *Streptococcus pneumoniae*, *Hemophilus influenzae*, *Klebsiella pneumoniae*, *Escherichia coli* were the most common microorganism found in the swab samples collected from the hospital. A routine bacterial surveillance of prevalent organisms and the study of the sensitivity patterns of the pathogens responsible for neonatal infection should be made an essential component for neonatal care. This information from many parts of the country will be important in policymaking on antimicrobial use not only locally but also internationally.

*Corresponding Author

Name: Anand Vijayakumar PR
Phone: +91-9443181573
Email: ootyand2004@gmail.com

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INTRODUCTION

Antibiogram is the most important activities performed by clinical microbiology that involves reporting of antimicrobial susceptibilities. Antibiogram helps in monitoring of antimicrobial resistance in ICU or ward specific data and inpatient versus outpatient data, etc. (Joshi, 2010).

Antibiograms are used by clinicians to find out local susceptibility rates and to monitor resistance trends over time within an institution. Antibiograms can also be used to compare susceptibility rates (Natarajan et al., 2016).

This study aims to determine if the current empiric treatment is adequate and effective. This will help reduce the risk of undertreatment or over the treatment of infections, both of which are associated with the emergence and increasing of resistance to antibiotics. The current data will be necessary in policy decisions and the development of treatment guidelines that can help to mitigate neonatal mortality. The antibiotic policy aims to prevent communicable diseases, decrease morbidity, and mortality due to antimicrobial-resistant infection, and to preserve the effectiveness of antimicrobial agents in treatment.

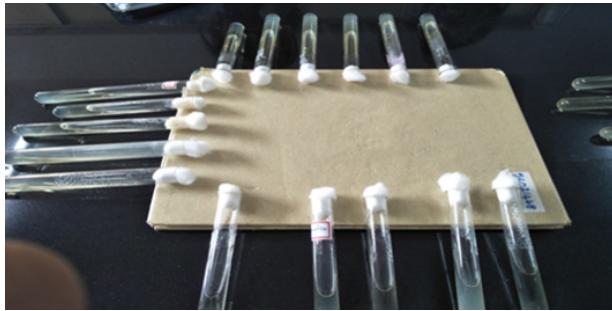


Figure 1: Swab samples incubated in nutrient broth

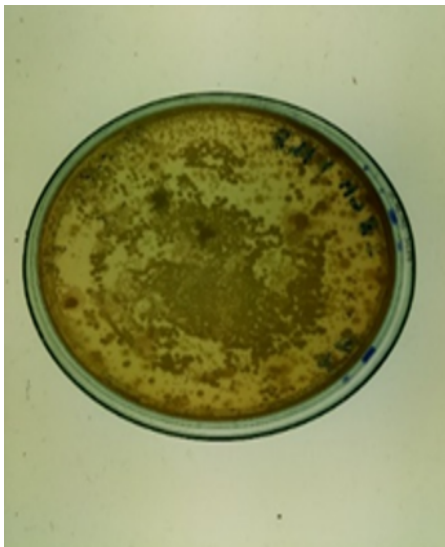


Figure 2: Growth obtained in Chocolate agar (*H. influenzae*)

MATERIALS AND METHODS

We conducted a prospective study for all wards present in hospitals. Swab samples were collected from the hospital for Antibiogram. The study was conducted at GHQH, Ooty, Nilgiris, Tamil Nadu, India. It is run by the State Government of Tamil Nadu. It has one labor ward, one ante-natal ward, and five post-natal wards with 120 beds. It was cho-



Figure 3: Growth obtained in Mac Conkey (*E.coli*)

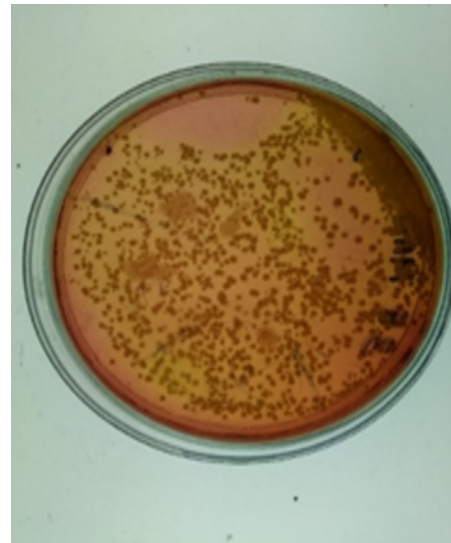


Figure 4: Growth obtained in Mannitol salt agar (*S.aureus*)

sen because it serves both low income and middle-income population and has a laboratory facility that was used for processing of the samples. The Institutional Ethical Committee approval was obtained from Institutional Ethics Committee, JSS College of Pharmacy, Rocklands, Ooty before the initiation of the study (Reference no: JSSCP/DPP/IRB/01/2017-18, Dated: 03.02.2018). Figures 1, 2, 3, 4 and 5.

Swab samples were collected from NICU, Laboratory, Blood bank, Labor room, Children Ward, ICU, Microbiology lab, AN Ward was isolated in nutrient agar and incubated. Bacterial growth were identified by gram staining, motility test, and antibiotics sensitivity, and resistance test was done.

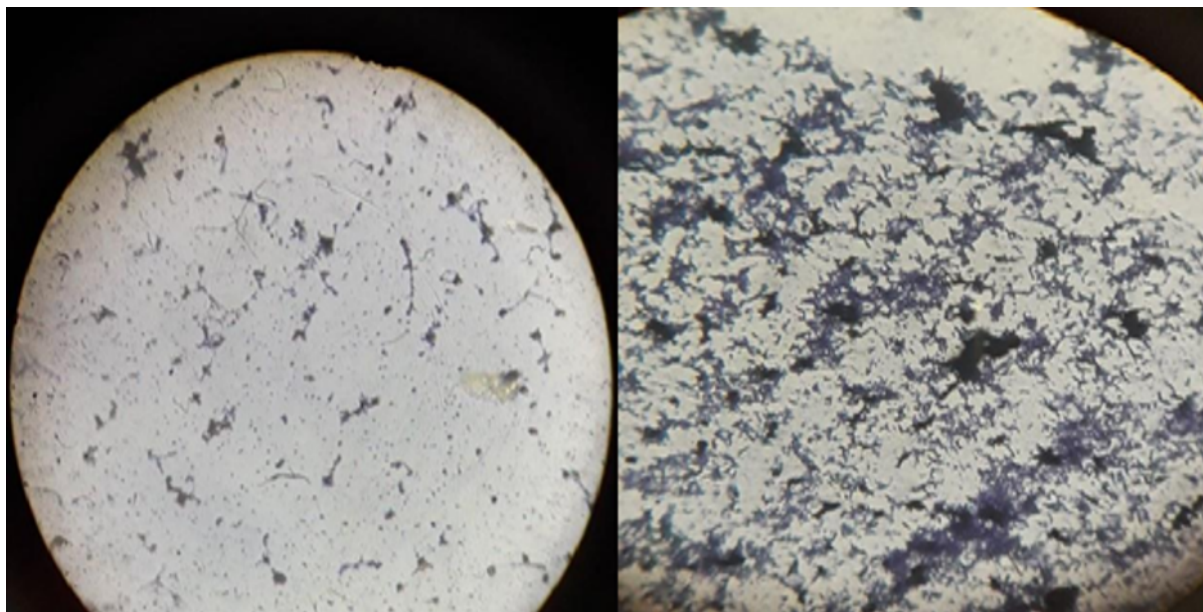


Figure 5: Rod shape and spherical shape is shown under a microscope

RESULTS AND DISCUSSION

This prospective study was conducted by collecting 72 swab samples from hospital.

Selective media that are used in swab sample to isolate the bacteria are,

1. Chocolate Agar
2. Mac Conkey Agar
3. Mannitol Salt Agar
4. EMB Agar

In Table 1, Samples tested from blood bank – Microscope (1A), Blood collecting monitoring (1B), REM1(1C), VDRL rotator (1D), Incubator (1E), REM 2 (1F), Floor (1G), ELIZA reader (1H), Walls (1I) and Centrifuge (1J). The growth were found in 9 samples except in the incubator, microorganisms present are *H.influenzae*, *S.pneumoniae*, and *S.aureus*.

In Table 2, Samples tested from children's ward - Patient cot (2A), Door window (2B), CH walls (2C), Incubator (2D), and Floor (2E). Growth was present in three agar except for EMB agar. Microorganisms present are *H.influenzae*, *S.epidermitis*, *Klebsiella pneumoniae*, *S.aureus*, and *E.coli*.

In Table 3, Samples tested from ICU- Pulse oximetry (3A), Window (3B), Ventilator (3C), Floor (3D), and Patient cot (3E). Growth was present in three agar except for EMB agar. Micro-organisms present are *H.influenzae*, *S.epidermitis*, *Klebsiella pneumoniae*, *S.aureus*, and *E.coli*.

In Table 4, Samples tested from DPH LAB- Incubator (4A), Floor (4B), Wall (4C), Distilled water (4D), an Auto analyzer (4E), Centrifuge (4F), Window (4G), Hot air oven (4H), Door (4I) and Microscope (4J). Growth was present in three agar except for EMB agar. Among these 10 samples, 3 samples were selected, and the Microorganisms present are *H.influenzae*, *S.epidermitis*, *Klebsiella pneumoniae*, *S.aureus*, *S.pneumoniae*, and *E.coli*.

In Table 5, Samples tested from Microbiology laboratory - Microscope(5A),Floor(5B), Window(5C), Working area(5D), Refrigerator 1(5E),Refrigerator 2(5F),Walls(5G),Deep freezer (5H), Incubator(5I) and Door (5J). Growth was present in three agar except for EMB agar. Microorganisms present are *H.influenzae*, *Klebsiella pneumoniae*, *S.aureus*, and *E.coli*.

In Table 6, Samples tested from OT- OT Table 2 (6A), Floor (6B), Instrument trolley (6C), Window (6D), Boyle's apparatus 2 (6E), Shadow lamp1 (6F), Shadow lamp 2 (6G), OT Table 1 (6H), Suction (6I) and Boyle's apparatus 1 (6J). Growth was present in three agar except EMB agar. Microorganisms present are *H.influenzae*, *S.pneumoniae*, *Klebsiella pneumoniae*, *S.aureus*, and *E.coli*.

In Table 7, Samples tested from Labor room – Ward door (7A), Incubator (7B), Table 7 (7C), Walls (7D), Labor instrument (7E), and Labor board (7F). Growth was present in three agar except EMB agar. Microorganisms present are *H.influenzae*, *S.pneumoniae*, *Klebsiella pneumoniae*, *S.aureus*, *E.coli*, and *Pseudomonas spp*.

In Table 8, Samples tested from Antenatal ward

Table 1: Testing for microorganism with swab samples collected from different places of Blood Bank their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Vancomycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
Blood bank(1A)	<i>H.influenzae</i>	S	S	S	S	MR	S	S
Blood bank(1B)	<i>S.pneumoniae</i>	R	R	MR	MR	MR	MR	S
Blood bank(1C)	<i>S.aureus</i>	S	S	S	S	S	S	S

S.pneumoniae showed resistance to both Ampicillin and Amikacin. *H.influenzae* showed mild resistance to Vancomycin, and *S.pneumoniae* showed mild resistance to Gentamicin, Vancomycin, and Piperacillin

Table 2: Testing for microorganism with swab samples collected from different places of Children Ward their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Vancomycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
CH(2A)	<i>H.influenza</i>	S	S	S	S	MR	S	S
	<i>S.epidermitis</i>	S	S	S	S	MR	MR	S
	<i>Kleb.pneumoniae</i>	Highly S	R	S	S	MR	S	S
CH(2B)	<i>E.coli</i>	Highly S	MR	S	S	R	MR	S
CH(2E)	<i>S. aureus</i>	S	S	S	S	S	S	S

Klebsiella pneumoniae showed resistance to Amikacin, and *E.coli* showed resistance to Vancomycin. *H.influenzae*, *S.epidermitis*, *Klebsiella pneumoniae* showed mild resistance to Vancomycin and Piperacillin. *E.coli* showed resistance to Amikacin and Piperacillin

Table 3: Testing for microorganism with swab samples collected from different places of Intensive Care Unit (ICU) their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Vancomycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
ICU(3A)	<i>H.influenza</i>	MR	S	S	S	MR	S	S
	<i>S.epidermitis</i>	S	MR	S	S	MR	MR	S
	<i>E.coli</i>	S	MR	S	S	R	MR	S
ICU(3D)	<i>E.coli</i>	S	S	S	S	S	S	S
ICU(3E)	<i>S.aureus</i>	S	R	S	S	MR	S	S

E.coli showed resistance to Vancomycin, and *S.aureus* showed resistance to Amikacin. *H.influenzae*, *S.epidermitis*, *E.coli*, and *S.aureus* showed mild resistance to Ampicillin, Amikacin, Vancomycin, and Piperacillin

room – Floor 1(8A), Wall 1(8B), Door and Window 1(8C), Door and Window 2(8D), Floor 2 (8E) and Wall 2(8F). Growth was present in three agar except EMB agar. Microorganisms present are *H.influenzae*, *S.aureus*, and *E.coli*.

In Table 9, Samples tested from NICU – Phototherapy 1(9A), Ventilator (9B), Wall 1(9C), Warmer 1(9D), Wall 2(9E), Warmer 2 (9F), Phototherapy 2(9G), and Floor 1 (9H), Window (9I) and Floor 2 (9J). Growth was present in three agar except EMB agar. Microorganisms present are *H.influenzae*,

Klebsiella pneumoniae, and *S.aureus*.

Microbiology, OT: Operation Theater, LR: Labor room, AW: Antenatal ward, NICU: Neonatal intensive care unit, S: Sensitive, MR: Mild Resistance, R: Resistance, *H.influenzae*- *Hemophilus influenzae*, *S.epidermitis* – *Staphylococcus epidermitis*, *Kleb. Pneumoniae* - *Klebsiella pneumoniae*, *S.aureus*- *Staphylococcus aureus*, *E.coli*- *Escherichia coli*, *S.pneumoniae*-*Streptococcus Pneumoniae***

Procedure for Gram Staining

Table 4: Testing for microorganism with swab samples collected from different places of DPH their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Van-comycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
DPH(4A)	<i>H.influenzae</i>	MR	S	S	S	MR	S	S
	<i>S.aureus</i>	S	S	S	S	S	S	S
	<i>Kleb.pneumoniae</i>	S	R	S	S	MR	S	S
DPH(4F)	<i>S.epidermitis</i>	S	S	S	S	S	S	S
	<i>S.pneumoniae</i>	R	R	MR	S	MR	R	S
DPH(4J)	<i>E.coli</i>	Highly S	MR	S	S	R	MR	S

Klebsiella pneumoniae showed resistance to Amikacin, *S.pneumoniae* showed resistance to Ampicillin, Amikacin, Piperacillin, and *E.coli* showed resistance to Vancomycin

H.influenzae and *Kleb.pneumoniae* showed mild resistance to Ampicillin and Vancomycin. *S.pneumoniae* and *E.coli* showed mild resistance to Gentamicin, Amikacin, Vancomycin, and Piperacillin

Table 5: Testing for microorganism with swab samples collected from different places of Microbiology Laboratory their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Van-comycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
Micro (5A)	<i>H.influenzae</i>	MR	MR	S	S	MR	S	S
	<i>S.aureus</i>	S	S	S	S	S	S	S
	<i>E.coli</i>	S	MR	S	S	R	MR	S
Micro(5J)	<i>Kleb.pneumoniae</i>	S	R	S	S	MR	S	S

E.coli and *Klebsiella pneumoniae* showed resistance to Vancomycin and Amikacin. *H.influenzae*, *E.coli*, and *Klebsiella pneumoniae* showed mild resistance to Ampicillin, Amikacin, Vancomycin, and Piperacillin

Table 6: Testing for microorganism with swab samples collected from different places of Operation Theatre (OT) their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Van-comycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
OT(6D)	<i>H.influenzae</i>	MR	MR	S	S	MR	S	S
	<i>S.aureus</i>	S	S	S	S	S	S	S
	<i>E.coli</i>	S	MR	S	S	R	MR	S
OT(6I)	<i>Kleb.pneumoniae</i>	S	R	S	S	MR	S	S
OT(6J)	<i>S.pneumoniae</i>	R	R	S	S	MR	MR	S

E.coli, *Klebsiella pneumoniae*, and *S.pneumoniae* showed resistant to Vancomycin, Ampicillin, and Amikacin. *H.influenzae*, *E.coli*, *Klebsiella pneumoniae*, *S.pneumoniae* showed mild resistance to Ampicillin, Amikacin, Vancomycin, Amikacin, and Piperacillin

Bacteria isolated were drop in their respective slides. After this, the slide was dried in the flame (Heat kill). After this, Crystal Violet was added and wait for 1 minute after a 1-minute wash with water. Add Gram's iodine and wait for 1 minute after a 1-minute wash with water. Add Alcohol (5%) and wait for 15 seconds, wash with water. Add Safranin and wait for 45 seconds, wash with water. Focus under the microscope (Acharya, 2015).

Hanging Drop Method

Depression slide was used to find the motility test in the hanging drop method. The microorganism was a drop in the middle of the coverslip. Each side of the coverslip, Vaseline, or liquid paraffin was drop. The coverslip was placed in the depression slide. The microorganism was observed under the microscope (Acharya, 2014).

Table 7: Testing for microorganism with swab samples collected from different places of Operation Theatre (OT) their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Vancomycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
LR(7C)	<i>H.influenzae</i>	S	S	S	S	MR	S	S
	<i>S.aureus</i>	S	S	S	S	S	S	S
	<i>E.coli</i>	MR	S	S	S	MR	S	S
LR(7D)	<i>Kleb.pneumoniae</i>	Highly S	R	S	S	S	S	S
LR(7E)	<i>S.pneumoniae</i>	R	R	MR	S	MR	R	S
	<i>Pseudomonas</i>	Highly S	R	S	S	R	MR	S

lebsiella pneumoniae, *S.pneumoniae* and *Pseudomonas* showed resistance to Amikacin, Ampicillin, Vancomycin and Piperacillin. *E.coli*, *H.influenzae*, *S.pneumoniae* and *Pseudomonas* showed mild resistance to Vancomycin, Ampicillin, Gentamicin and Piperacillin

Table 8: Testing for microorganism with swab samples collected from different places of Antenatal Ward (AW) their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Vancomycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
AW(8A)	<i>H.influenzae</i>	MR	S	S	S	MR	S	S
		S	S	S	S	S	S	S
	<i>S.aureus</i>	S	MR	S	S	R	S	S
	<i>E.coli</i>							

E.coli showed more resistance to Vancomycin. *H.influenzae* and *E.coli* showed mild resistant to Ampicillin, Vancomycin, and Amikacin

Table 9: Testing for microorganism with swab samples collected from different places of Neonatal Intensive Care Unit (NICU) their resistance and sensitivity patterns towards various antibiotics

Name of the Ward	Species identified	Ampicillin 10mcg	Amikacin 30mcg	GM 10 mcg	GM 120mcg	Vancomycin 30mcg	Piperacillin 100mcg	Meropenem 10mcg
NICU (9A)	<i>H.influenzae</i>	MR	S	S	S	MR	S	S
		S	S	S	S	S	S	S
	<i>S.aureus</i>							
NICU (9B)	<i>Kleb.pneumoniae</i>	S	R	S	S	MR	S	S
NICU (9E)	<i>S.epidermitis</i>	S	MR	S	S	MR	MR	S

Klebsiella pneumoniae showed resistance towards Amikacin. *H.influenzae*, *Klebsiella pneumoniae*, and *S.epidermitis* showed mild resistance towards Ampicillin, Vancomycin, Amikacin, and Piperacillin.

**CH: Children ward, DPH: Director of Public Health, ICU: intensive care unit

When compared with different swab samples collected in the hospital, we have found various types of microorganisms present in hospitals of different wards. Microorganisms found from swab samples are *H. influenzae*, *S. aureus*, *Klebsiella pneumoniae*, *E. coli*, *S. pneumoniae*, *S. epidermitis*, and *Pseudomonas spp.* The most common pathogens found from various wards and rooms are *H. influenzae*, *S. aureus*, *E. coli*, and *Klebsiella pneumoniae*. All the organisms showed absolute sensitivity mostly to Meropenem, Gentamicin, Ampicillin, and Piperacillin. Ampicillin showed highly sensitivity to organisms like *E. coli*, *Klebsiella pneumoniae*, *S. pneumoniae*, and *Pseudomonas*. Amikacin showed resistance towards to *S. pneumoniae*, *Klebsiella pneumoniae*, *S. aureus*, and *Pseudomonas*. Vancomycin 30mcg, Piperacillin 100mcg, Amikacin 30mcg, and Ampicillin 10mcg showed mild resistance to *S. pneumoniae*, *Klebsiella pneumoniae*, *S. aureus*, *S. epidermitis*, *E. coli*, *Pseudomonas* and *H. influenzae*.

Antibiogram includes laboratory testing for sensitivity and resistance of an isolated bacteria to different common use antibiotics. Susceptibility patterns of antimicrobial is becoming less predictable. Due to the irrational use of antibiotics, resistance to antibiotics increases (Brook and Long, 2018). Bacteria resistance has become a threatening infection worldwide in both hospital and community settings (Namratha et al., 2015).

Antibiograms are used to define a rational use and selection of antimicrobial therapy in hospitals for treating patients. It is made for multiple purposes and to assist clinicians to follow proper empirical therapy (Joseph et al., 2011).

CONCLUSIONS

From the collected swab samples, *H. influenzae*, *S. aureus*, *E. coli*, and *Klebsiella pneumoniae* are the most common pathogens implicated in hospitals from different wards. All the organisms showed sensitivity towards antibiotics, but Gentamicin, Piperacillin, Meropenem, and Ampicillin showed higher sensitivity compared to other antibiotics. Antibiogram will be useful for the incorporation of informations and patients data. Antibiotic susceptibility may help in overcoming the problems of antibiotics use and to follow proper antibiotic policies.

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