

# International Journal of Advanced Research in Biological Sciences

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## Research Article

### Etiological profile of pathogenic isolates in intensive care unit of tertiary care hospitals

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#### Abstract

The study of the microbial interactions with animals becomes the prime importance for diagnosis and treatment of dreadful diseases in clinical industry. Although the primary interest is in diseases caused by these interactions, it must also be appreciated that microorganisms play a critical role in human survival. The normal commensal population of microbes participates in the metabolism of food products, provides essential growth factors, protects against infections with highly virulent microorganisms, and stimulates the immune response. In the absence of these organisms, life as we know it would be impossible. Nosocomial infection is defined as an infection which develops 48 hours after hospital admission or within 48 hours after being discharged. The objectives were to assess the frequency of nosocomial infection in patients admitted to intensive care unit (ICU) and to determine the etiological factors in such patients. An understanding of medical microbiology requires knowledge not only of the different classes of microbes but also of their propensity for causing disease. The most frequent site of infection was respiratory tract (23%) and urinary tract (23%). *Pseudomonas* (P.), *Klebsiella* (K.), *Escherichia* (E.) *coli*, *Staphylococcus* sp, *Acinetobacter* and *Candida* spp. were the commonest organisms.

**Keywords:** microbial interactions, normal commensal, respiratory tract, urinary tract.

## Introduction

The microbial flora in and on the human body is in a continual state of flux determined by a variety of factors, such as age, diet, hormonal state, health, and personal hygiene. An understanding of medical microbiology requires knowledge not only of the different classes of microbes but also of their propensity for causing disease. A few infections are caused by strict pathogens and others are caused by opportunistic pathogens. A hospital-acquired infection is usually one that first appears three days after a patient is admitted to a hospital or other health care facility. The microbial population that colonizes the human body is numerous and diverse. While a patient is admitted to the hospital for treatment of other conditions, nosocomial infections may be acquired from other patients, hospital staff, contaminated objects or solutions, or from the patient himself (such as transfer from one site of the body to another). Most common nosocomial infections are

surgical wound infections, urinary and respiratory tract infections, and bacteremia (bloodstream infections). Many of these infections are by antibiotic-resistant bacteria, known as superbugs, and can often have serious consequences for the individual and the hospital community. Nosocomial infection is also known as hospital or healthcare-acquired infections (Ingrid Koo, 2009). Historically, *Staphylococci*, *Pseudomonads*, and *Escherichia coli* have been the nosocomial infection; nosocomial pneumonia, surgical wound infections, and vascular access-related bacteremia have caused the most illness and death in hospitalized patients; and intensive care units have been the epicenters of antibiotic resistance. Aging of our population and increasingly aggressive medical and surgical interventions, including implanted foreign bodies, organ transplantations, and xeno-transplantation, create a cohort of particularly susceptible persons. Nosocomial infection rates in adult

and pediatric ICUs are approximately three times higher than else were in hospitals. The sites of infection and the pathogens involved are directly related to treatment in ICUs. The source of the infecting organism may be exogenous - from another patient or a member of the hospital staff, or from the inanimate environment in the hospital; or it may be **endogenous** from the patient own flora which at the time of infection may include organisms brought into the hospital at admission and certain others acquired subsequently.

For an infection to occur in the hospital the prerequisites are:

- (a) A susceptible host.
- (b) A microbe capable of producing an infection.
- (c) An environment that is congenial for the multiplication of the microbe.

### Objective

This was a prospective study carried out at the tertiary care hospitals in and around Bangalore over a period of three months. A total of 50 isolates were obtained from 45 patients admitted to the ICU wards of these hospitals. The aim was to study nosocomial infections among surgical patients.

- To study about the pathogenic organisms which are most prevalent in the ICU wards.
- This study was planned to delineate the occurrence, microbiology of the infectious organisms in the tertiary care hospitals.

Nosocomial infection is defined as an infection which develops 48 hours after hospital admission or within 48 hours after being discharged (Jan Muhammad Shaikh *et al.*, 2008). The intensive care unit (ICU) often is called the epicenter of infections, due to its extremely vulnerable population (reduced host defences deregulating the immune responses) and increased risk of becoming infected through multiple procedures and use of invasive devices distorting the anatomical integrity-protective barriers of patients (intubation, mechanical ventilation, vascular abscesses, etc.) (Marwick C *et al.*, 2009). Patients admitted to the ICU have been shown to be at particular risk of acquiring nosocomial infection with a prevalence rate as high as 30% (Craven DE *et al.*, 1988). The risk of nosocomial infection in ICU is 5–10 times greater than those acquired in general medical and surgical wards (Vincent JL *et al.*, 1995). Nosocomial infection is an important health-care problem (Wilkins E *et al.*, 1996). According to WHO

manifestation, 5-10% of hospitalized patients of developed countries and about 25% of developing countries were affected by a nosocomial infection in 2005 (Dunbar E *et al.*, 1996).

### Materials and methods

#### Sample collection

The clinical samples were collected from the patients in and nearby to Bangalore using pre-sterilized robust, leak-proof, sterile containers. Precautional measures were taken to minimize the contamination. The clinical sample was mixed well and processed on the same day. Different samples like pus samples, endotracheal samples, ear swab, throat samples, tissue samples, sputum samples, urine samples, CVP samples were collected.

#### Identification of isolates

The bacterial isolates obtained after the primary screening was maintained in pure culture on Nutrient Agar Media plates. All the media plates were incubated at 37°C until used. Study of colony morphology of the isolated cultures was carried out. Gram's staining was performed. Further motility test was performed followed by culturing the isolates on culture media and finally biochemical test in order to differentiate one pathogen among the other. Biochemical characterization of the isolated colonies was carried out using standard protocols. Identification was carried out according to Bergey's Manual (7<sup>th</sup> Ed.). All the media used during the course of the study were obtained from Hi media laboratories Pvt. Limited (A- 406, Bhaveshwar plaza, Mumbai-400086, India).

Different isolates obtained from the ICU patients were collected in sterile glassware and were transported to the laboratory for further processing in the transport media. When some time may elapse before the sample is examined, and especially where delicate pathogens, may be present, it is advantageous to place the swab in a transport medium, such as Stuart's medium, which preserves the stability of the pathogens. The medium is non-nutrient, because the less delicate commensal bacteria present in the specimen would outgrow the pathogens in a nutrient medium.

#### Maintenance of pure culture

The colonies obtained were analyzed for colony morphology and sub cultured in Nutrient Agar plates

incubated at 30-35 for 42 hours and then stored at a refrigeration temperature of 4 C for further use.

## Result and discussion

### Central venous catheter sample

Central venous pressure is considered a direct measurement of the blood pressure in the right atrium and vena cava. A central venous catheter can be used to give chemotherapy drugs. Some CVCs have more than one lumen (opening) and can be used to give more than one drug at a time. Central venous catheters can be left in place for weeks to months. It is an intravenous line that is used for giving the patient fluids and / or medications.

3 isolates were obtained from CVP (central venous catheter pipe) sample out of which two isolates were of *Klebsiella* and one isolate of *Acinetobacter* based upon the biochemical tests.

### Ear swab sample

Ear swabs were taken from the external auditory meatus mainly in 3 suspected conditions, acute otitis media, chronic suppurative otitis media and otitis externa. The provisional clinical diagnosis will indicate the different organisms likely to be present.

The isolates obtained from the ear swab sample were found to be one isolate of *Staphylococcus* and one isolate of *Pseudomonas* based upon the biochemical tests.

### endotracheal sample

The trachea, commonly known as the windpipe, is a tube about 4 inches long and less than an inch in diameter in most people. The trachea begins just under the larynx (voice box) and runs down behind the breastbone (sternum). Infections of the lower respiratory tract are the leading cause of cause of mortality worldwide. A catheter that is inserted into the trachea through the mouth or nose in order to maintain an open air passage or to deliver oxygen or to permit the suctioning of mucus or to prevent aspiration of the stomach contents is called endotracheal tube.

10 isolates were obtained from endotracheal sample, out of which three isolates were of *Staphylococcus* four isolates of *Pseudomonas* and one isolate of

*Acinetobacter*, *Klebsiella* & *Bacillus* based upon the biochemical tests.

### Pus sample

Pus is an exudate, typically white-yellow, yellow, or yellow-brown, formed at the site of inflammation during infection. An accumulation of pus in an enclosed tissue space is known as an abscess, whereas a visible collection of pus within or beneath the epidermis, it causes lumps as a pustule or pimple. Pus consists of a thin, protein-rich fluid, known as liquor puris and dead leukocytes from the body's immune response (mostly neutrophils).

8 isolates were obtained from Pus sample out of that two isolates of *Staphylococcus*, two isolates of *Streptococcus* and one isolate of *Citrobacter*, *Klebsiella*, *Pseudomonas* & *Providentia* based upon the biochemical tests.

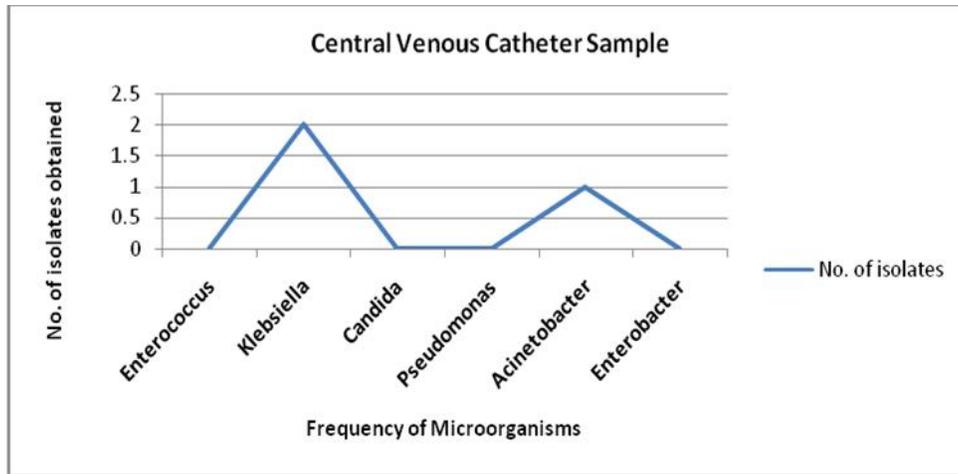
### Sputum sample

Sputum is a mucousy substance (consisting of cells and other matter) that is secreted into the airways of the respiratory tract. The material from lower respiratory infections most commonly submitted for bacteriological examination is sputum, a mixture of bronchial secretion and inflammatory exudates coughed up into the mouth and expectorated. There are difficulties in collecting a suitable sample and in interpreting the result of its culture.

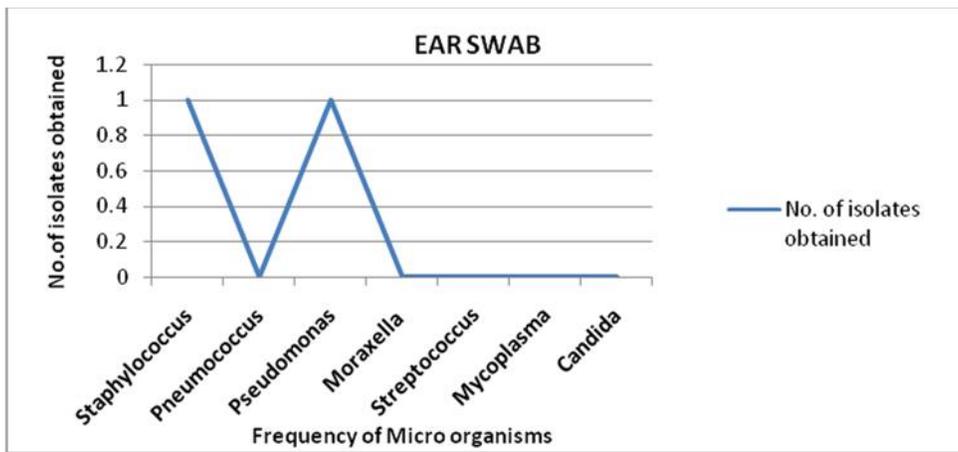
13 isolates were obtained from Sputum sample amongst them three isolates were of *Citrobacter*, two isolates of *Pseudomonas*, two isolates of *Candida*, two isolates of *Streptococcus*, two isolates of *Acinetobacter*, and one isolate of *Actinomyces* & *E. coli*. The *Actinomyces* was grown in selective media known as Glycerol Asparagine Agar.

### Throat sample

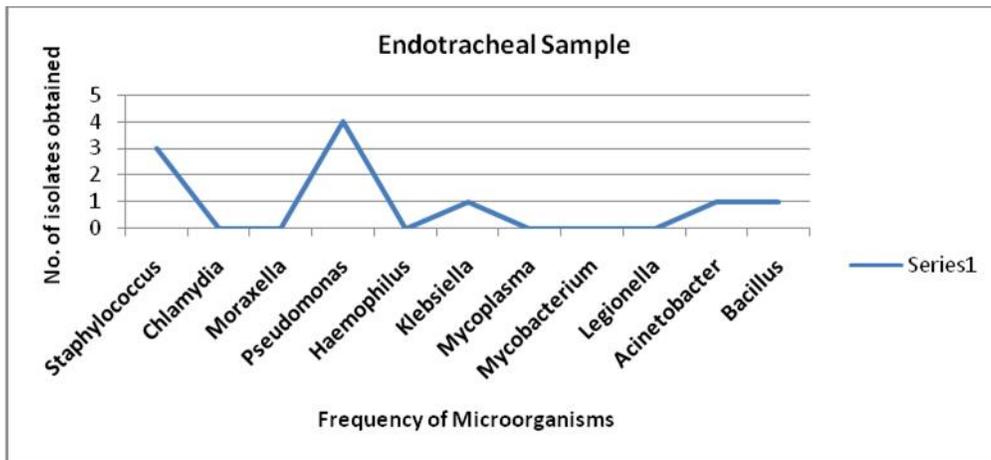
The commonest respiratory infections are localized in the oropharynx, nasopharynx and nasal cavity, causing sore throat, nasal discharge and often fever, but the throat pathogens may also spread to infect the larynx, causing hoarseness; the middle ear, causing otitis media with ear ache, eye, causing conjunctivitis or keratitis etc. The upper respiratory tract is frequently involved in wider or generalized infections such as whooping cough and measles.



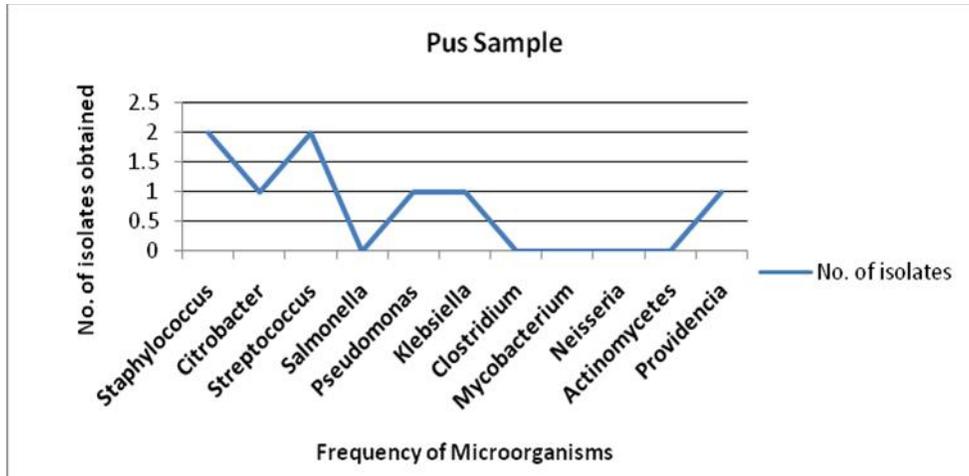
**Fig: 1** Frequencies of Micro Organisms Obtained from CVP Sample



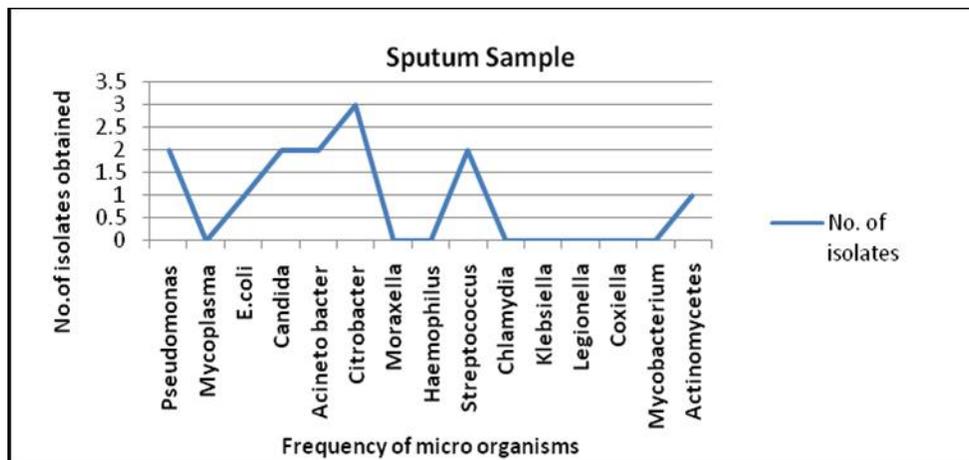
**Fig: 2** Frequencies of Micro Organisms Obtained from Ear Swab



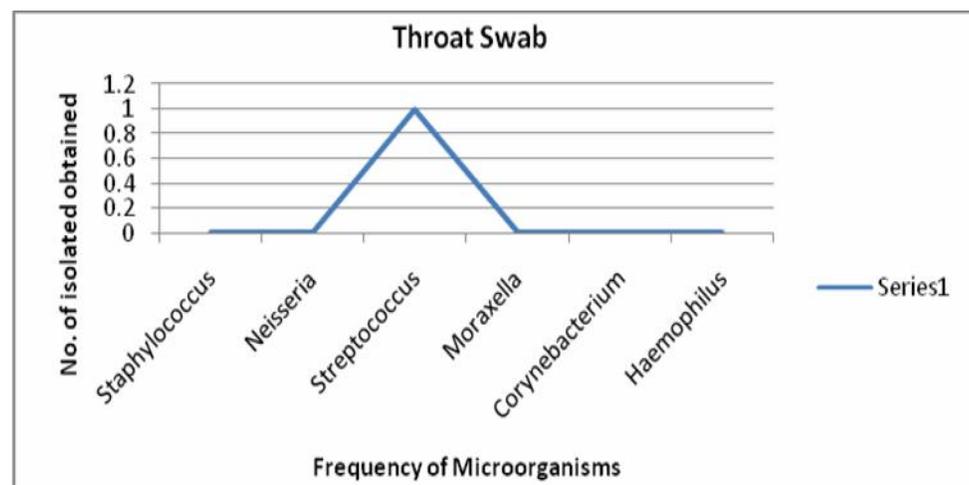
**Fig: 3** Frequencies of Micro Organisms Obtained from ET Sample



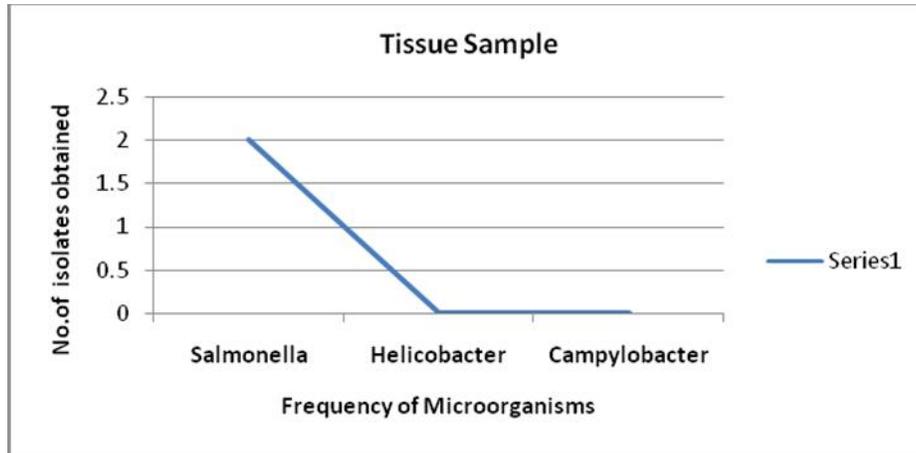
**Fig: 4** Frequencies of Micro Organisms Obtained from Pus Sample



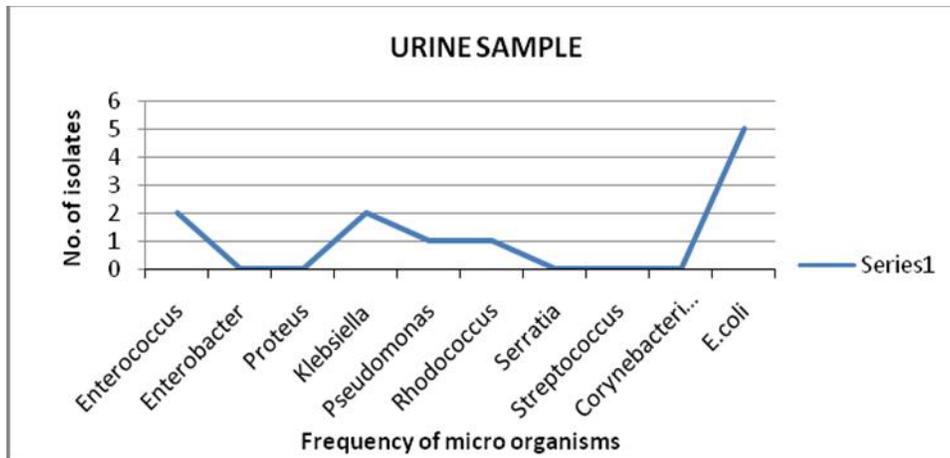
**Fig: 5** Frequencies of Micro Organisms Obtained from Sputum Sample



**Fig: 6** Frequencies of Micro Organisms Obtained from Throat Sample



**Fig: 7** Frequencies of Micro Organisms Obtained from Tissue Sample



**Fig: 8** Frequencies of Micro Organisms Obtained from Urine Sample

1 isolate was obtained from Throat sample and that was found *Streptococcus* sp.

### Tissue culture

Tissues are collected for histopathology, parasitology, biochemistry, immuno-histochemistry; cultures and demonstration of DNA. For histopathology samples can be preserved by fixation, for all other techniques, fresh samples are required. There are several methods that can be employed in the sampling tissues some of them are: Brush biopsy cytology, Curettings, Punch biopsy, Brush biopsy cytology etc. One of the most common tissue sampling method is Endoscopy.

2 isolates were obtained from Tissue sample and was found to be gram negative bacilli i.e., *Salmonella* of based upon the biochemical tests.

### Urine sample

Urine is a typically sterile liquid by-product of the body secreted by the kidneys through a process called urination and excreted through the urethra. Urine has a long, rich history as a source for measuring health and well-being and remains an important tool for clinical diagnosis. The clinical information obtained from a urine specimen is influenced by the collection method, timing and handling.

11 isolates were obtained from Urine sample, among them five isolates of *E.coli*, two isolates of *Enterococcus*, two isolates of *Klebsiella* and one isolate of *Rhodococcus* & *Pseudomonas* were obtained based upon the biochemical tests.

## Summary

Nosocomial infections become prominent in surgical wards because of surgical intervention and operative procedures and furthermore hospitals particularly acute care units, surgical and medical units are important breeding ground for the development and spread of antibiotic resistant bacteria. A total of 50 isolates were recovered from 45 patients enrolled over a period of three months out of which most were bacterial and very few were fungal. The samples from the patients were collected once they admitted to the ICU section within 48 hours. In this study, the samples were collected, processed by staining method, by inoculating them into the selective media, followed by biochemical test and finally antibiogram was done in order to obtain the resistance pattern of the organisms present in the ICU wards. Different organisms were obtained from different patients from different samples such as tissue, ear, CVP, sputum, endotracheal, throat, pus, urine. It was revealed from the study that, the organisms obtained from patients were mostly the normal flora of the body which became pathogenic due to the decrease in the immune level of the patients, due to age variation etc. Gram-negative organisms are the predominant pathogens causing infections in surgical patients. So for proper management of critically ill patients and patients undergoing various operative procedures and other medical interventions, hospital antibiotic policies need frequent revisions. The aim of this study therefore is to determine the most common bacterial pathogens associated with the disease in the intensive care unit of the tertiary care hospitals.

## References

- Anathanarayan., and Pankir. 2006. Test book of microbiology 7th edition, 356pp.
- Aneja, K.R., 2008. *Experiments in Microbiology, Plant Pathology and Biotechnology (Fourth Edition)*. pp 253-274.
- Craven, D.E., Kunches, L.M., Lichtenberg, D.A., Kollisch, N.R., Barry, M.A., and Heeren, T.C.1988. Nosocomial infection and fatality in medical and surgical intensive care unit patients. *Arch Intern Med*; 148(5):1161–8.
- <http://infectiousdiseases.about.com/bio/Ingrid-Koo-Ph-D-50107.htm>.
- Jan, M. S., Bikha, R. D., Syed, Zulfiqar. A. S., and Tauseefullah, A. I. B. 2008. Frequency, pattern and etiology of nosocomial infection in intensive care

- unit: an experience at a tertiary care hospital. *J Ayub Med Coll Abbottabad*; 20(4).
- Mackie and McCartney. 2004. 4<sup>th</sup> edition. *Practical Medical Microbiology*. pp 58-129.
- Marwick, C., and Davey, P. 2009. The holy grail of infectious risk management in hospital? *Curr Opin Infect Dis*, 22:364-369.
- Vincent, J.L., Bihari, D.J., Suter, P.M., Bruining, H.A., White, J., and Nicolas, C. 1995. The prevalence of nosocomial infections in intensive care units in Europe. Results of the European Prevalence of Infection in Intensive Care (EPIC) Study. EPIC International Advisory Committee. *JAMA*; 274:639-44. Comment in: *JAMA* 1996; 275:362.
- Wilkins, E., Mandal, B., and Dunbar, E.1996. *Infectious Disease*. Oxford: Blackwell; 58th ed.