

The Study of Bacterial Ecology in Intensive Care Unit of Mohammed V Military Teaching Hospital, Rabat, Morocco

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ABSTRACT

The study of the bacterial ecology in a surgical intensive care unit has a huge interest for the fight against the infections. the aim of this work is to analyze the epidemiological and the bacteriological profiles of infections, also to identify the risk factors and others influence the prognosis, finally to propose the measures of prevention. during one year at the bacteriology unit of mohammed V military teaching hospital of Rabat city in Morocco this prospective study was giving: the results of the total samples about the surgical intensive care unit patients. The characteristics of the patients “having infectious state” in intensive care unit: the middle age is 45,5 years old, the majority of cases correspond to male people, the high blood pressure (35,29%) and diabetes (23,53%) are the most frequent breeding . the severe cranial trauma corresponds to the most cases (16, 33%), secondly, we find the polytrauma 14, 28%. the infection incidence was considered as 6, 10%. the gram-negative bacilli are the most observed (69, 23%), secondly, we find the gram-positive cocci 23,10%. the klebsiella pneumoniae is the most frequently identified species (25%), is the most frequently isolated on catheters samples, it has an important susceptibility to imipeneme. the acinetobacter baumannii is present in the second row (13,46%), having a susceptibility to colistine. the escherichia coli (9, 62%) and staphylococcus coagulase negative (9,62%) are both in the third row, this one is the most frequently isolated species in blood cultures and presents a neatly susceptibility to teicoplanine and vancomicine. in the fourth row we find enterococcus faecalis (7, 69%), corresponds to the second germ isolated from protected bronchial specimens, practically found in the almost specimens, and presents a high susceptibility to ampicilline, vancomycine and teicoplanine. the mortality dealing with infections is 55, 56%, while the mortality happened to “sepsis states” is 20%; the male people are frequently affected (73%). a global strategy of prevention must be established for fighting infections and making better the prognosis.

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Introduction

The study of bacterial ecology is of major interest in the management of infections in the intensive care unit, due to the severity of the pathology responsible at admission, the frequent presence of previous chronic diseases, a major antibiotic consumption linked to the frequency of nosocomial infectious pathologies and a high rate of patients carrying multi-resistant bacteria (MRM) [1]. The aim of this work is to update the bacterial ecology of nosocomial infections and to follow the evolution of the antibiotic susceptibility profile of

bacteria in a context of changing epidemiology of antibiotic resistance. This study was extended over a one-year period and involved the bacteriological analysis of 75 samples and the collection of the necessary information on 49 patients who are the subject of this study. The patients studied corresponded to all the patients who presented an infectious state [2]. At the beginning or after their stay in intensive care. In the latter case, they must meet the diagnostic criteria for nosocomial infection [1,3]. Bacterial ecology studies must be

regularly produced at least once a year [4], allowing the implementation of an infection surveillance strategy [5,6].

Tools and Methods

This is two forms of studies, a prospective study within the Bacteriology Department of the Mohammed V Military Training Hospital (HMIMV) in Rabat, over a period of one (from January 2016 to December 2017), the results of which are the subject of a descriptive study, and a retrospective study carried out secondarily, from which all the necessary clinical information was gathered concerning the patients (hospitalized in the HMIMV's surgical intensive care unit, in whom the bacteriological samples initially analyzed were taken.

The diagnosis of an infection or an infectious state corresponding to the different forms; acquired (nosocomial infection) or imported (Community) in hospital is often difficult to establish because of the great heterogeneity of clinical presentations, and the difficulty in defining its nosological framework. For this reason, an effort has been made to standardize a consensual definition, leading to a widely used consensus concerning the definition of the main syndromes corresponding to the successive phases of aggravation of the infection and the resulting inflammatory response [2,7]. An infectious state or infection may be Probable [2], when there is clinical, biological or sometimes radiological evidence of an infection, but without microbiological confirmation, can be confirmed by evidence of the germ from a specimen. Specific codified criteria [1,7] were developed for the positive diagnosis of different infectious states in the intensive care unit, depending on the infected site, and based on clinical, radiological and biological evidence. The bacteriological and clinical data thus collected were reproduced on Microsoft Excel spreadsheets and adapted to SPSS 18 (Statistical Package for Social Sciences) for statistical analysis.

Results

The characteristics of the patients who presented an infectious state: the average age is 45.5 years, male sex predominates, hypertension (35.29%) and diabetes (23.53%) being the most frequent pathological terrain, isolated cranial traumas are the most frequent (16.33%), ahead of polytrauma (14.28%), and hemorrhagic strokes (12.24%). The incidence of infections was 6.10%. The total number of patients studied recalling the, is 49 patients, having benefited from a total number of 90 specimens. There were multiple specimens corresponding to a single patient, 10 patients having received multiple specimens (at least 2 specimens), representing 20.40% of all patients studied. The incidence rate of bacterial infections: about 6.10%.

A- Study of Infections related to the sampling site

A total of 90 samples were analyzed, 38 samples were found to be positive, a positivity rate of about 42.22%. Approximately 21.05% of the positive samples were blood cultures, cytobacteriological examinations of urine, catheter samples and protected distal swabs (PDS) each accounted for 18.42%. Table 1 shows the distribution of positive specimens (infections) by specimen site.

Table 1 . Distribution of Positive Samples by Site

Sampling Site	Percentage%
B-C (Blood-Culture)	21,10
ECBU (Cytobactériological examination of urine)	18,42
KT (Catheters)	18,42
PDS (Protected distal swabs)	18,42
DPUS (Deep Pus)	10,53
SPUS (Superficial Pus)	10,53
UK (Urinary catheter)	2,63
TOTAL	100

B- Bacterial epidemiology

The distribution of bacteria according to gram staining shows that about 69.23% of the total bacterial species isolated are Gram-negative Bacilli (GNB), 23.10% are Gram-positive Cocci (GPC), and only 7.70% are Gram-positive Bacilli (GPB), while the distribution of bacteria according to group shows that Fermenting Bacteria (FB) represent about 53.85%, Non-Fermenting Bacteria (NFB) represent about 23.10% and COCCI represent about 23.05%.

The study of the bacteriological profile according to species has isolated a total number of 52 bacterial species, of which *Klebsiella Pneumoniae* occupies the first place among the species isolated with a rate of approximately 25%, *Acinetobacter baumannii* occupies the second position with a rate of approximately 13.46% and *Staphylococcus aureus* and *Escherichia coli* share the third place with a rate of approximately 9.62%. Table 2 shows the distribution of all bacteria isolated according to their species.

Table 2.: Distribution of all isolated bacteria according to their species

Species	Number	Percentage%
KP (<i>Klebsiella pneumoniae</i>)	13	25
AB (<i>Acinetobacter baumannii</i>)	7	13,46
NCS (Negative coagulase <i>staphylococcus</i>)	5	9,62
EC (<i>Escherichia coli</i>)	5	9,62
EF (<i>Enterococcus faecalis</i>)	4	7,69
PA (<i>Pseudomonas aeruginosa</i>)	3	5,77
ECL (<i>Enterobacter cloacae</i>)	3	5,77
CS (<i>Corynebacterium species</i>)	3	5,77
SA (<i>Staphylococcus aureus</i>)	2	3,85
AS (<i>Acinetobacter species</i>)	2	3,85
PM (<i>Proteus mirabilis</i>)	2	3,85
SS (<i>Staphylococcus saprophyticus</i>)	1	1,92
SM (<i>Serratia marcescens</i>)	1	1,92
ASpp (<i>Actinomyces Spp</i>)	1	1,92
TOTAL	52	100

The study of the Bacteriological Profile by specimen type showed that 11 bacterial species were isolated in blood cultures, with coagulase-negative *Staphylococcus* occupying the first place with a rate of approximately 27.27%, while *Klebsiella pneumoniae* shares the second place with *Acinetobacter baumannii* with a rate of approximately 18.18%. Polymicrobial character was observed in approximately 37.5% of cases. Table 3 shows the distribution of bacterial species isolated from all blood cultures.

Among the 11 bacterial species isolated, *Acinetobacter baumannii* was predominant in about 36.36% of cases, *Pseudomonas aeruginosa* was in second place with a rate of 27.27% and *Klebsiella pneumoniae* was in third place. The polymicrobial character is observed in 57.14% of cases. Table 4 shows the distribution of bacterial species isolated at the PDS.

Catheter samples allowed the isolation of 10 bacterial species, with an observed polymicrobial rate of approximately 42.86%. *Klebsiella pneumoniae* is the most isolated bacterial species in 30% of cases.

Among all the positive samples obtained from ECBU (cytobacteriological examination of urine), 9 bacterial species were identified, a predominance of *Klebsiella pneumoniae* was noted with a rate of approximately 55.55%, while *Escherichia coli* is in second place with a rate of approximately 22.22%. Polymicrobial character is observed in 28.57%.

C- Bacterial sensitivity to antibiotics

- Sensitivity profile of bacterial groups

The main antibiotics used by the bacteriology laboratory of HMIMV Rabat to determine the rate of sensitivity (% S), intermediate sensitivity (% IS) and resistance (% R) of the different bacteria isolated are summarized in table 5, note that the rate of cases in which an antibiotic donated is not tested (NT%) are always mentioned in our study.

The Fermenting Bacteria (FB), present a high rate of sensitivity to IMPENEME (75.83%), to AMIKACIN (57.77%), to COLISTINE (62.77%), to GENTAMICIN (61.38%), and to NETILMICIN (52.77%), Non-Fermenting Bacteria (NFB) have a high sensitivity rate to COLISTINE (57.14%) and AMIKACIN (74.74%), and a resistance rate to IMPENEME reaching 40.47%, while Cocci: Having shown 100% sensitivity to VANCOMYCIN and TEICOPLANIN.

- Sensitivity profile of bacterial species (in order of frequency)

Klebsiella pneumoniae; has a high sensitivity to IMPENEME (80%), AMIKACIN (70%), and COLISTINE (60%). Figure 1 shows the activity of Antibiotics against *Klebsiella pneumoniae*. *Acinetobacter baumannii*; has a high sensitivity to COLISTINE (71.43%), and AMIKACIN. Figure 2 explains the activity of Antibiotics against *Acinetobacter baumannii*. *Escherichia coli*; has a total sensitivity to FOSFOMYCIN, AMIKACIN, and COLISTIN. High sensitivity to IMPENEME (75%), and to NETILMICIN (50%). *Staphylococcus coagulase-negative*; full sensitivity to TEICOPLANINE, and VANCOMYCIN, and high sensitivity to TETRACYCLINE (80%), FUSIC ACID (80%), CHLORAMPHENICOL (60%), and FOSFOMYCIN (60%). *Enterococcus faecalis*; is 100% sensitive to AMPICILLIN, TEICOPLANINE, and VANCOMYCIN.

Table 3 . Distribution of Bacterial Species isolated at blood-cultures

Type of sampling	Bacterial species	Number	Percentage %	Polymicrobism rate %
Blood-Cultures	NCS (Negative coagulase <i>Staphylococcus</i>)	3	27,27	37,5
	KP (<i>Klebsiella pneumoniae</i>)	2	18,18	
	AB (<i>Acinetobacter baumannii</i>)	2	18,18	
	SA (<i>Staphylococcus aureus</i>)	1	9,09	
	ECL (<i>enterobacter cloacae</i>)	1	9,09	
	CS (<i>Corynebacterium species</i>)	1	9,09	
	AS (<i>Acinetobacter species</i>)	1	9,09	
TOTAL		11	100	

Table 4 . Distribution of Bacterial Species identified in protected distal swabs

Type of sampling	Bacterial species	Number	Percentage	Polymicrobism Rate %
PDS	AB (<i>Acinetobacter baumannii</i>)	4	36,36	57,14
	EF (<i>Enterococcus faecalis</i>)	3	27,27	
	KP (<i>Klebsiella pneumoniae</i>)	2	18,18	
	CS (<i>Corynebacterium species</i>)	1	9,09	
	PA (<i>Pseudomonas aeruginosa</i>)	1	9,09	
TOTAL		11	100	

Table 5 . Legend of the antibiotics tested by the HMIMV bacteriology laboratory in Rabat

AMX	Amoxicillin	LEV	Levofloxacin
TIC	Ticarcillin	MXF	Moxifloxacin
PIP	Piperacillin	NOR	Norfloxacin
AMC	Amoxicillin + clavulanic acid	OFX	Ofloxacin
TCC	Ticarcillin + Ac clavulanique	PEF	Pefloxacin
TZP	Piperacillin + Tazobactam	NA	Nalidixic Acid
KF	Cefalotine	MIN	Minocyclin
CXM	Cefuroxime	TE	Tetracyclin
FOX	Cefoxitine	E	Erythromycin
MA	Cefamandole	MY	Lincomycin
TRX	Ceftriaxone	DA	Dalacine
CTX	Cefotaxime	TEC	Teicoplanin
FEP	Cefepim	VA	Vancomycin
ATM	Aztreonam	RIF	Rifampicin
CFS	Cefsulodine	FD	Fusidic acid
PYC	Ceftazidime	CT	Colistine
MOX	Latamoxef	C	Chloramphénicol
IMP	Imipenem	FOS	Fosfomycin
ERP	Ertapenem	SMX	Sulfaméthoxazole / Trimethoprim
OX	Oxacillin	SP	Spiramycin
PG	Penicillin G	DOX	Doxycycline
AK	Amikacin	MTZ	Metronidazole
GN	Gentamicin	NS	Nistatine
K	Kanamycin	NTF	Nitrofurantoin
NET	Netilmicin	LZ	Lizenolide
TOB	Tobramycin	CEFP	Cefpirome
CIP	Ciprofloxacin	FLM	Flumeric acid

Significant sensitivity to GENTAMYCIN (75%), CHLORAMPHENICOL (50%), RIFAMPICIN (50%), and FOSFOMYCIN (50%). *Pseudomonas aeruginosa*; is 100% sensitive to IMPENEME, with significant sensitivity to CEFTAZIDIME (66.66), TOBRAMYCIN (66.66), AMIKACIN, FOSFOMYCIN (66.66), and TAZOBACTAM+PIPERACILLIN. *Enterobacter cloacae*; is 100% sensitive to IMPENEME, and FOSFOMYCIN, with high sensitivity to AMIKACIN, NETILMYCIN, TAZOBACTAM+PIPERACILLIN, and COLISTIN, and finally *Corynebacterium* species; is 100% sensitive to TEICOPLANIN, VANCOMYCIN, and GENTAMYCIN.

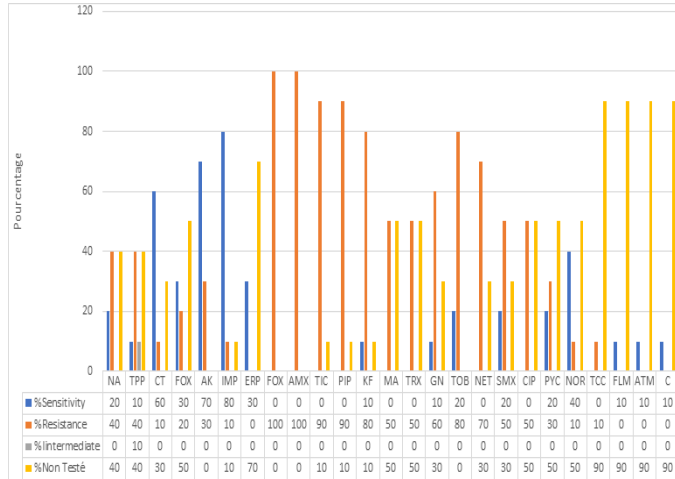


Figure 1. Activity of Antibiotics against *Klebsiella pneumoniae*

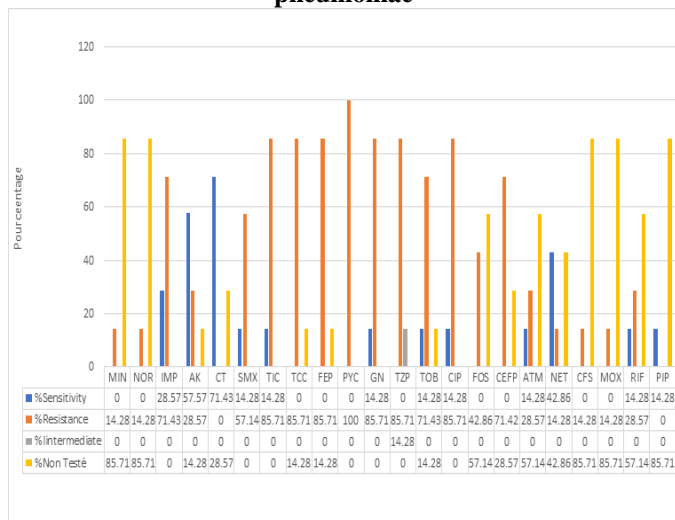


Figure 2. Activity of Antibiotics against *Acinetobacter Baumannii*

Discussion

A recent prospective international study conducted in 28 intensive care units (Multicenter Study) in eight different countries estimated the incidence of infections at 21.1%. 28% of these infections met the criteria for sepsis, 24% for severe sepsis, and 30% for septic shock (18% remained unclassified) [2,8]. There is, however, great variability in distribution between countries, as shown in the study by Vincent et al [2,9]. On the other hand, 20% of patients infected on admission or during their stay, without any criteria of severity, progress to severe sepsis or septic shock within 10 days [2]. Another American study, conducted in three hospitals, evaluated the frequency of progression from sepsis to severe sepsis in 9% of patients and from severe sepsis to septic shock in 3% [2,10]. In-hospital mortality related to sepsis remains

high, between 15 and 60%, and rises if sepsis develops during hospitalization [2]. According to our study, the rate corresponding to the incidence of infections during 2009 is about 6.10%. This rate is lower than that observed in France according to Brun-Buisson in 2001 [11]: 14.1%. The incidence rate of nosocomial infections remains difficult to establish with certainty because it depends on the conditions of collection and recruitment of services, inclusion and exclusion criteria. In many studies carried out in this field, the first factor of variation is the type of definitions used [12-14], these definitions must be as close as possible to those given by the CDC, and adapted in France to the context of resuscitation by the Réanis group [14-16]. The frequency of isolation of a germ and its level of resistance varies according to regions, from one hospital to another, from one unit to another and within the same unit from one period to another. Only multi-center studies can give a fairly accurate idea of the frequency of germs isolated from surgical resuscitation patients with their level of resistance [14-17]. In France, the 1996 national survey on the prevalence of infections revealed that Gram-negative Bacilli (BGN) represent 53% while Gram-positive Coccus (CGP) represent 33% [18]. According to another study carried out in 2003 at the Ibn Rochd University Hospital in Casablanca [19]: BGN were 73.5%, and CGP represent 26.5% of the germs isolated, which is close to our study (BGN: 69.23% and CGP: 23.10%). However, in recent years, it has been observed that CGP predominates. The SCOPE project revealed that PMCs were isolated in 64% of 10617 bacteremia episodes [20], the same for Struelens: PMCs represent the predominant category [21], other studies have shown similar results: according to Vincent 2006, PMCs predominate [2,9]. According to a study at the Ibn Rochd university hospital[19], *Acinetobacter* predominates among BGN, which is not the case in our study where *Klebsiella pneumoniae* is in first place with a rate of about 25%, while *Acinetobacter Baumannii* is in second place in our series with a rate of about 13.46%. According to another study conducted in the intensive care unit of the Mohammed V Military Training Hospital in Rabat [22], the bacteriological profile of blood culture isolates during the period 2002-2005 was as follows: *Acinetobacter baumannii* 13.64%, *Staphylococcus epidermidis* 12.6%, *Staphylococcus aureus* 11.9%. *Pseudomonas aeruginosa* occupies the last rank 7%. According to our study, the blood culture profile was found to consist of: Coagulase-negative *Staphylococcus* 27.27%, *Klebsiella pneumoniae* 18.18%, *Acinetobacter baumannii* 18%, *Staphylococcus aureus* 9.09%. The constant presence of *Staphylococcus* and *Acinetobacter baumannii* in blood cultures was then observed, but a predominance of BGN 63.63% compared to CGP 36.37% was observed in our study. While some French multi-center studies dealing with laboratory-based bacteremia surveillance [23], and other American studies have shown a predominance of CGP [24]. *Klebsiella pneumoniae* is the bacterium most implicated in urinary tract infections 55.55%, and catheter-related infections 30%. It causes up to 5% of community-acquired urinary tract infections and 9% of nosocomial infections [25]. *Acinetobacter baumannii* is the first bacterium to be isolated from protected distal specimens (36.36%). *Klebsiella pneumoniae*: belonging to the group of fermenting species is the most frequent and most isolated germ in the different types of samples in our series. It presents a clear sensitivity to imipenem, amikacin and colistin, an important resistance to betalactamines, the same results were obtained according to a study carried out in HMIMV's medical

intensive care unit [26], this can be explained - according to a French multicenter study - by a large diffusion of betalactamase producing strains (35 centers) in 1996 [27]. *Acinetobacter baumannii*: It is one of the non-fermenting bacilli which, according to our studies, represent 13.46% of the total bacterial ecology of surgical resuscitation at HMIMV in Rabat. The incidence of *Acinetobacter baumannii* infections has increased considerably over the last 30 years, particularly in high-risk wards (intensive care units) [28]. This bacterium is endowed with a great capacity to persist in the environment, which contributes to the difficulty of eradicating its outbreaks. In recent years, antibiotic resistance of *Acinetobacter baumannii* has been steadily increasing and intra-hospital epidemics due to multi-drug resistant strains are regularly reported [24]. Imipenem resistance in *Acinetobacter baumannii* is a phenomenon increasingly reported in our hospital, according to a study carried out at the HMMV on the "study of the sensitivity of isolated strains" [29], and another carried out in the intensive care unit [22]: There is still a moderate to moderate resistance to all the antibiotics tested, including Imipenem except colistin, in our series *acinetobacter baumannii* was resistant to Imipenem with a resistance rate of about 71.43%, this rate was close to that obtained in a study carried out at the CHU Sahloul in Tunisia, where the percentage rate of resistance rose from 44% in 2002 to 51.5% in 2004 [30]. According to another study carried out in HMMV medical intensive care during 2009 [26], the rate of resistance to imipenem was 48%, indicating the presence in these bacteria of an extremely vast and diverse arsenal to counteract the antibiotic action and a very wide range of combinations of resistance and opportunities for dissemination. Phenotypic and molecular studies looking for imipenem resistance mechanisms in these strains have demonstrated the co-existence of non-enzymatic resistance mechanisms (membrane impermeability) and enzymatic mechanisms (production of oxacillinases with carbapenemase activity) [personal data]. In our series, coagulase-negative *Staphylococcus* was 100% sensitive to teicoplanin and vancomycin. According to the NNIS (National Nosocomial Infections Surveillance) study covering the period 1986-1989, coagulase-negative *Staphylococcus* represents 9% of the bacteria responsible for nosocomial infections, all sites combined, and a high sensitivity to these two antibiotics [25]. Most of the bacteria found in our series show moderate to significant resistance to all the antibiotics tested. This may reflect their nosocomial nature. Among all the bacteria found, two bacterial species were isolated: *Proteus Mirabilis*, which was sensitive to all the antibiotics tested. This germ was identified from a single mono-microbial sample from PUSPRO (deeper), and *Staphylococcus saprophyticus*, which was only resistant to Fosfomycin, which may testify to their communal nature. In order to make clinical studies relevant and comparable, the respect of diagnostic criteria and definitions is crucial, due to the great heterogeneity of the syndrome and the affected population, it is important, especially for clinical studies, to identify a target population based mainly on clinical characteristics and parameters influencing the fate of patients. This explains the increasing importance given to the so-called "generalist" severity scores: SAPS, Acute Physiology and Chronic Health Evaluation (APACHE), Mortality Probability Model (MPM), Sepsis - Related Organ Failure score (SOFA), Logistic Organ Dysfunction System (LODS), or "organ-specific" scores which have been shown to correlate with mortality: Glasgow

Coma Scale for Neurological Failure, Acute Injury Kidney Injury Network (AKIN) for Renal Failure, and to the Chronology of the disease including its duration and delay, and finally to the differentiation and recognition of other associated infectious causes. The nature of the pathogen and the site of infection have an influence on mortality, but other factors are taken into account to determine the prognosis, whether they are related to the characteristics of the germ in question, the infected site or the particularities of the host itself. Our study attempted to establish an epidemiological and bacteriological framework of infection in the ICU based on the monitoring of specimen results obtained for each patient. Nevertheless, the difficulty to define the infected subjects from a clinical point of view "Infectious states" and the non-recognition of the diagnostic criteria considered for each patient, limited us to determine with certainty the exact number of patients considered infected.

Conclusion

Nosocomial infections are more frequent in the ICU than in other areas of care due to the critical condition of patients and the invasive nature of life support techniques. Their surveillance is essential to quantify the risk of infection and monitor its evolution. They constitute a real problem with multiple consequences in terms of morbidity, which is assessed by the lengthening of the length of hospitalization, mortality and overcost. The agents involved are often multi-resistant. Hence the need for a global strategy for the prevention and control of nosocomial infection, which requires close collaboration between epidemiologists, clinicians, bacteriologists, hygienists and the healthcare team. Nevertheless, in order to control the risk of infection, such a strategy can only be conceived within the framework of a global approach to the quality of care in intensive care units.

Current state of knowledge on the subject

- A study of bacterial epidemiology in several intensive care units in Europe and North America has made it possible to improve the management of nosocomial infections and prevent antibiotic resistance.

- Several strategies to prevent nosocomial infections in intensive care units include studies of microbial ecology.

- Nosocomial infections are a real problem with multiple consequences in terms of morbidity, which is assessed by the lengthening of hospital stays, mortality and overcrowding.

Contribution of our study to knowledge

- Latest study of bacterial epidemiology carried out at the surgical intensive care unit level in order to update the bacterial ecology of nosocomial infections and monitor the evolution of the antibiotic sensitivity profile of isolated bacteria.

- *Klebsiella Pneumoniae* is the most frequent and isolated bacterium with a clear sensitivity to Imipenem, while *Acinetobacter Baumannii* is the second most frequent bacterium with a clear sensitivity to Colimycin.

- The need for a global strategy to prevent and fight against nosocomial infection is only conceivable within the framework of a global approach of quality care in intensive care units.

Consent

As per international standard or university standard, patient's consent has been collected and preserved by the authors.

Ethical Approval

It is not applicable.

Competing Interests

Authors have declared that no competing interests exists.

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