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EPIDEMIOLOGICAL ANALYSIS OF BACTERIAL STRAINS INVOLVED IN HOSPITAL INFECTION IN A UNIVERSITY HOSPITAL FROM BRAZIL

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SUMMARY

Hospital infections cause an increase in morbidity and mortality of hospitalized patients with significant rise in hospital costs. The aim of this work was an epidemiological analysis of hospital infection cases occurred in a public University Hospital in Rio de Janeiro. Hence, 238 strains were isolated from 14 different clinical materials of 166 patients hospitalized in the period between August 1995 and July 1997. The average age of the patients was 33.4 years, 72.9% used antimicrobials before having a positive culture. The most common risk conditions were surgery (19.3%), positive HIV or AIDS (18.1%) and lung disease (16.9%). 24 different bacterial species were identified, *S. aureus* (21%) and *P. aeruginosa* (18.5%) were predominant. Among 50 *S. aureus* isolated strains 36% were classified as MRSA (Methicillin Resistant *S. aureus*). The Gram negative bacteria presented high resistance to aminoglycosides and cephalosporins. A diarrhea outbreak, detected in high-risk neonatology ward, was caused by *Salmonella* serovar Infantis strain, with high antimicrobial resistance and a plasmid of high molecular weight (98Mda) containing virulence genes and positive for R factor.

KEYWORDS: Hospital infection; Antimicrobial resistance; *Salmonella* outbreak

INTRODUCTION

Hospital Infections (HI) are a public health problem, mainly, because they increase morbidity and mortality of hospitalized patients, time of hospital stay and costs of treatment (JARVIS, 1987).

In developed countries HI ranges from 5 to 10% of hospitalizations (BERGOGNE-BÉRÉZIN, 1995). It is estimated that in the United States from 25,000 to 100,000 deaths are caused directly by HI, with correspondent costs of over 7.5 billion dollars a year (WENZEL, 1994; SMITH & DOEBBELING, 1996).

Some risk conditions, which are likely to cause HI, have been reported. Some of them are: rise in the number of surgeries, use of catheters, mechanical ventilation, immunosuppressors and new bacterial strains resistant to antimicrobials (AL ORAINEY *et al.*, 1989; SARTOR *et al.*, 1995; KOELEMAN *et al.*, 1997).

The microorganisms related to HI have varied in the last five decades, due to environmental factors and selective pressure of antimicrobial use. Thus, in the 50s the first outbreaks of *S. aureus* resistant to penicillin happened between 1960-1980, *S. aureus* resistant to oxacillin emerged as well as the Gram negative fermenters, which produce β -lactamases. After 1980 there were reports of Gram negative non-fermenters, *Candida sp.* and some types of virus (JARVIS, 1987, AL-ORAINY *et al.*, 1989,

COTTON *et al.*, 1989, BERGOGNE-BÉRÉZIN, 1995; SARTOR *et al.*, 1995, MONNET *et al.*, 1997).

This study reports the frequency of the microorganisms associated with HI in a public University Hospital in Rio de Janeiro, Brazil. It emphasizes the distribution of the microorganisms in several wards, anatomic sites, resistance to antimicrobials and epidemiological data of the patients infected.

MATERIAL AND METHODS

Description of the Hospital and Population

Bacterial strains were isolated from patients in different hospital wards of the Hospital Universitário Gaffrée & Guinle (HUGG) – Universidade do Rio de Janeiro (UNI-RIO), Brazil. This hospital provides secondary and tertiary assistance to patients of different ages, from several places in Rio de Janeiro City and State. The data collected on each infection included the date, site of infection and patient demographic characteristics.

Bacterial Strains and Susceptibility Testing

A total of 238 bacteria strains isolated from 14 different anatomic sites (Table 2) were obtained from patients with infection. The criteria

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for defining nosocomial infection followed the National Nosocomial Infection Surveillance System (NNISS) according recommendations of Ministry of Health-Brazil, 1994. NNISS defines a nosocomial infection as any infection that is not present or incubating at the time the patient is admitted to the hospital, so was considered IH as being any infection occurrence after 48h of hospitalization. Each patient's record was analysed criteriously, and based on clinical aspects, laboratorial data, type of infection and clinical specimen, the strain was classified as representative of infection or not. Surgical site infection were classified as infection occurrence within 30 days of the operation. Neonates infections were classified as any infection acquired after birth. All ConS strains were reported as the cause of a bloodstream infection only if the patient had fever, chills or hypotension and had no clinical evidence of sepsis at another site. The culture and identification followed the rules of BALOWS *et al.* (1991). The Gram-negative identification was completed by the Crystal System of identification for fermenters and non-fermenters (BBL/Becton-Dickinson). The antimicrobial susceptibility test was carried out through disk diffusion method in agar-disk of CECON, complying with NCCLS (National Committee for Clinical Laboratory Standards) recommendations (1995). Quality control was carried out using standard strains of *Escherichia coli* (ATCC 25922), *P. aeruginosa* (ATCC 27953) and *S. aureus* (ATCC 25923).

Plasmid Profile Analysis

Plasmid Profile analysis was performed through the alkaline lysis method of BIRNBOIM & DOLY (1979), modified by SAMBROOK *et al.* (1989). Samples of 15µl plasmid preparation were loaded into wells with 0.8% agarose gel of 7.5 cm length and run at 40V for 2 hours on a horizontal electrophoresis apparatus with a constant power source. Gels were stained with ethidium bromide and visualized on an UV transilluminator. Photographs were taken with a Polaroid Type 667 film. The plasmidial DNA of *Escherichia coli* R861 and V517 (THRELLFALL *et al.*, 1986; MACRINA *et al.*, 1978) were included as molecular controls.

Determination of the presence of R factors

Cultures of *Salmonella* serovar Infantis and of the receptor strains *Escherichia coli* K12 F, Lac⁺ and Nal^r (originally from the Biophysics Institute, Federal University of Rio de Janeiro) were incubated at 1:10 inoculum proportion at 37 °C for 18 hr. Transconjugants were plated onto Agar McConkey, with the antimicrobial drugs added individually at the following concentrations: ampicillin, 10 mcg/mL – gentamicin – 10mcg/mL; ceftriaxone – 16 mcg/mL. All plates contained nalidixic acid at the concentration of 30 mcg/mL.

The growth of lactose-positive colonies (transconjugants) indicated the transfer of resistance markers. This result was confirmed by an antimicrobial susceptibility test and plasmid profile analysis of the transconjugants, considering the original pattern of the corresponding donor colony.

Epidemiological analysis of patients

For each HI case an epidemiological report was done. It contained patients' data. These data were saved in an EXCEL 5.0 program (Microsoft) and analyzed later in Epi-Info Version 6.04b - January-1997.

RESULTS

According to the epidemiological report, several parameters were analysed. General features of 166 patients studied can be seen in Table 1. The average age was 33.4, and average number of days of hospitalization before the HI was 25.4 days. 72.9% of the patients used prophylactic or therapeutic antimicrobials before presenting a positive culture. In the case of polymicrobial infections, an average of 2.9 strains per anatomic site was detected. The risk conditions which were mostly related to the HI process were surgery (19.3%), HIV infection (18.1%), lung disease (16.9%) and prematurity (12.0%).

A total of 28 different types of antimicrobials (Table 2) were used prior to the positive culture, with a predominance of amikacin (11.9%), ampicillin (11.6%), gentamicin (11.0%) and ceftriaxone (10.0%).

24 different species were identified (Table 3), with higher frequency of *S. aureus* (21.0%) followed by *P. aeruginosa* (18.5%); Coagulase-negative staphylococci (9.7%) and *K. pneumoniae* (8.8%). The 50 *S. aureus* strains were mainly distributed in Internal Medicine Wards (IMW) and General Surgery Wards (GSW). 17 (38.6%) strains of *P. aeruginosa* were from IMW and 9 (20.4%) from GSW. The other species were uniformly distributed by other wards. We detected an outbreak of *Salmonella* serovar Infantis in the Neonatology Intensive Care Unity.

Table 1
Epidemiological data of the 166 patients with nosocomial infections caused by 238 agents

<i>Epidemiological Data</i>	Total Number of Patients -166
Mean Age ± SD	33.4 ± 27.8
Male	105 (63.3%)
Female	61 (36.7%)
Mean of hospitalization days before HI presentation	25.4 ± 27.9
Number of patients in use of antibiotics before positive culture	121 (72.9%)
Mean of antibiotic use days before positive culture	15.9
Mean number of infection /patient	1.6
Number of polymicrobial infections cases	37 (22.3%)
Mean number of strains /polimicrobial infections cases (*)	2.9
Risk conditions of 166 patients	
Surgical case	32 (19.3 %)
HIV infection	30 (18.1%)
Lung disease	28 (16.9%)
Prematurity	20 (12.0%)
Diabetes	11 (6.6%)
Renal distress	10 (6.0%)
Cancer	10 (6.0%)
Others	19 (11.4%)
Patient data not found	6 (3.6%)

SD = Standard deviation; (*)—Total of 107 isolated strains

Table 2

Frequency of antimicrobial use before the positive culture isolation

Antimicrobials	Frequency of use*	% (N=121)
Amikacin	38	11.9
Ampicillin	37	11.6
Gentamicin	35	11.0
Ceftriaxone	32	10.0
Cephalotin	25	7.8
Oxacillin	23	7.2
Trimethoprim-sulfametoxazole	21	6.6
Ceftazidime	17	5.3
Vancomycin	16	5.0
Cefotaxime	14	4.4
Penicillin	11	3.4
Cefoxitine	11	3.4
Ciprofloxacin	9	2.8
Cephalexin	4	1.2
Clindamicin	4	1.2
Metronidazole	4	1.2
Chloramphenicol	3	0.9
Norfloxacin	2	0.6
Erythromycin	2	0.6
Rifampicin	2	0.6
Imipenem	1	0.3
Tobramycin	1	0.3
Tetracycline	1	0.3
Nitrofurantoin	1	0.3
Cefuroxime	1	0.3
Amoxacillin	1	0.3
Rovamicin	1	0.3
Sulfadiazine	1	0.3

*- Many patients were treated with more than one drug at the same time.

The number of bacterial strains isolated according to the type of clinical material cultured is shown in Table 4. *S. aureus* was the predominant bacterium in surgical wound, blood, vascular catheter and cutaneous lesions. *P. aeruginosa* were most frequently isolated in surgical wound abdominal secretions and enteric tract, Coagulase- negative staphylococci (CoNS), *K. pneumoniae* and *A. baumannii* were frequently isolated from blood.

Considering the resistance to antimicrobial agents, 36% of the *S. aureus* were resistant to oxacillin. The resistance of Gram negatives to the antimicrobials is shown in Table 5. We observed a high percentage of resistance to aminoglycosides (gentamicin and amikacin) and 3rd generation of cephalosporins (Ceftriaxone and Ceftazidime). There was a high percentage of *P. aeruginosa* (59.5%) resistant to carbenicillin and of *A. baumannii* resistant to ciprofloxacin, in spite of the latter being a controlled antimicrobial. Imipenem was the most active agent against *K. pneumoniae*, the others *Enterobacteriaceae* (*E. coli*, *E. cloacae*, *P. vulgaris*, *M. organii*, *P. rustigianii* and *S. marcescens*) were more resistant.

An outbreak of *Salmonella* serovar Infantis, occurred between 05/April -20/April/1996 affected nine hospitalized children in the high risk nursery (Table 6). These were studied in more detail. All of them had

severe disease: six had respiratory problems and three had low body weight. The antimicrobial resistance and plasmidial DNA profiles of all the strains were identical, with only one plasmid (98 Mda). In the determination of the presence of R factors, all of the strains transferred the resistance plasmid to the standard strain recipient *E. coli* K12.

DISCUSSION

The HUGG provides assistance to adults, children and neonatology. It is, also, a reference hospital for some diseases such as: acquired immunodeficiency syndrome, diabetes and cancer. In this way, a large part of the patients, due to their underlying diseases are more especially susceptible to acquired hospital infections.

This bacteriological study, complemented with information about patients in hospital, had some aspects. The *S. aureus* (21%) was the predominant pathogen (Tables 3, and 4). SADER *et al.* (1999), in a multicentric study, related similar result in blood and surgical wounds infections.

In Table 1, it is interesting to point out the average number of days of hospitalization (25.4 days) before the diagnosis of HI, a long time of hospitalization, as well as, the long time of previous use of antimicrobials (15.9 days). These factors permit the emergence of multiresistant bacteria. MANRIQUE & GALVÃO (1996), reported that the use of prophylactic or therapeutic antimicrobials, may result in the spread of multiresistant strains in the hospital. Another aspect is that risk conditions or underlying diseases may have favored the occurrence of HI in some hospital areas, mainly in surgery wards and intensive care units. The Table 3 shows that wards SW, IMW, NICU and PICU sum up 36% of the places studied, according to SARTOR *et al.* (1995).

Polimicrobial infection (22.3% of the cases) represented a challenge in establishing proper treatment and emphasized the importance of obtain cultures for every bacterial infection in hospitalized patients before starting an empiric treatment. DUNCAN *et al.*, (1994), mention that some infections demand empiric antimicrobial therapy; however, this type of therapy should be guided by previous study *in vitro* susceptibility of the most frequent the HI bacterial strains of higher frequency in the hospitals.

Regarding the frequency of the microorganisms, *S. aureus* and *P. aeruginosa* sum up 39.5% of the total isolates; Coagulase- negative staphylococci (CoNS), *K. pneumoniae*, *E. coli* and *A. baumannii* sum up 32.4%. The other 28.2% were represented by other 18 bacterial species. These microorganisms have been described in the literature as the main causes of HI in hospitals that provide tertiary assistance to HIV patients and high risk newborns (AYLIFFE, 1997; JARVIS, 1987; BERGOGNE-BÉRÉZIN, 1995; SARTOR *et al.*, 1995; MONNET *et al.*, 1997; STROUD *et al.*, 1997; SADER *et al.*, 1999). It is also important to point out that the distribution of microorganisms mostly involved in HI is directly related to the origin (wards) of the patients and type of clinical material. The knowledge of the hospital indigenous flora isolated from each anatomical sites allows a better choice of empirical treatment.

The level of multiresistance of the species of the KES group (*Klebsiella sp.*, *Enterobacter sp.* and *Serratia sp.*) is worrying, since these microorganisms are rather frequent, distributed in several wards of the hospital. The multiresistance in this group is related to the

Table 3
Frequency of bacterial strains according the ward

Microrganism	GSW*	IMW	NICU	PW	ICU	PICU	Total (%)
<i>Staphylococcus aureus</i>	12	25	4	1	2	6	50 (21.0)
<i>Staphylococcus coag. neg.</i>	7	3	4	1	1	7	23 (9.7)
<i>Streptococcus sp.</i>	3	1	1	-	1	-	6 (2.5)
<i>Klebsiella pneumoniae</i>	5	4	2	3	2	5	21 (8.8)
<i>Klebsiella oxytoca</i>	-	-	1	-	-	-	1 (0.4)
<i>Escherichia coli</i>	12	3	-	-	1	2	18 (7.6)
<i>Enterobacter aerogenes</i>	3	1	-	-	-	1	5 (2.1)
<i>Enterobacter cloacae</i>	1	-	1	-	1	4	7 (2.9)
<i>Enterobacter sakazakii</i>	-	-	-	-	-	1	1 (0.4)
<i>Enterobacter sp.</i>	1	-	-	-	-	1	2 (0.8)
<i>Proteus mirabilis</i>	4	2	-	-	-	1	7 (2.9)
<i>Proteus vulgaris</i>	2	-	-	-	1	-	3 (1.3)
<i>Providencia rustigianii</i>	-	-	1	-	-	-	1 (0.4)
<i>Morganella morganii</i>	5	1	-	-	-	2	8 (3.4)
<i>Serratia marcescens</i>	1	2	-	-	-	1	4 (1.7)
<i>Citrobacter freundii</i>	2	-	-	-	-	-	2 (0.8)
<i>Pseudomonas aeruginosa</i>	9	17	4	3	7	4	44 (18.5)
<i>Burkholderia cepacia</i>	-	-	-	-	-	2	2 (0.8)
<i>Pseudomonas putida</i>	-	1	-	-	-	-	1 (0.4)
<i>Stenotrophomonas maltophilia</i>	1	3	-	-	-	-	4 (1.7)
<i>Acinetobacter baumannii</i>	4	5	3	-	1	2	15 (6.3)
<i>Flavobacterium odoratum</i>	1	-	-	-	-	-	1 (0.4)
<i>Salmonella serovar Infantis</i>	-	-	9	-	-	-	9 (3.8)
Non fermenters Gram negatives	1	1	-	-	1	-	3 (1.3)
Total	74	69	30	8	18	39	238

GSW* = General Surgery Wards; IMW = Internal Medicine Wards; NICU = Neonatology Intensive Care Unity; PW = Pediatric Ward; ICU = Intensive Care Unity; PICU= Pediatric Intensive

Table 4
Number of bacterial strains isolated according to the site of infection

Microrganism	Site of Infection														CNS	TOTAL
	SW	BL	VC	UR	CT	AS	LR	ET	EA	EY	OR	UT	BO			
<i>Staphylococcus aureus</i>	15	10	8	3	4	1	3	1	2	-	2	-	1	-	50	
<i>Coagulase Neg. Staphylococci</i>	4	6	5	1	3	3	-	-	-	1	-	-	-	-	23	
<i>Klebsiella pneumoniae</i>	1	6	3	2	2	3	2	-	1	-	-	1	-	-	21	
<i>Escherichia coli</i>	7	2	-	1	1	4	-	1	-	-	-	1	-	1	18	
<i>Enterobacter sp.</i>	3	5	1	2	2	2	-	-	-	-	-	-	-	-	15	
<i>Morganella morganii</i>	1	2	-	1	1	3	-	-	-	-	-	-	-	-	8	
<i>Acinetobacter baumannii</i>	2	7	3	2	-	1	-	-	-	-	-	-	-	-	15	
<i>Pseudomonas aeruginosa</i>	10	2	3	6	2	3	9	2	3	3	1	-	-	-	44	
OTHERS	10	3	2	1	1	8	4	10	1	1	2	-	-	1	44	
TOTAL	53	43	25	19	16	28	18	14	7	5	5	2	1	2	238	

SW= Surgical Wound; BL = Blood; VC = Vascular catheters; UR= Upper Respiratory tract; CT=Cutaneous; AS = Abdominal secretion; LR= Lower respiratory tract; ET = Enteric tract; EA = ear; EY = eye; OR= Oropharyngis; UT = Urinary tract; BO = Bone; CNS = Central nervous system.

Table 5
Percentage of resistance to antimicrobial agents in Gram negatives strains

Microorganism	Antimicrobial													
	AMP	CPL	CFO	CRX	CRO	CAZ	CAR	IMP	CIP	GEN	AMI	TET	CLO	TSX
<i>K. pneumoniae</i>	100	76	14	76	57	-	-	0	5	57	57	52	48	52
<i>K. oxytoca</i>	100	100	0	100	100	-	-	0	0	100	100	0	100	100
<i>E. coli</i>	82	67	11	39	11	-	-	5	11	11	11	59	39	41
<i>E. aerogenes</i>	100	100	80	80	60	-	-	0	0	40	20	60	60	60
<i>E. cloacae</i>	100	100	100	100	86	-	-	14	29	86	86	86	100	57
<i>E. sakazakii</i>	100	100	100	100	100	-	-	0	0	100	100	0	100	100
<i>Enterobacter sp.</i>	100	50	100	50	100	-	-	0	50	100	100	100	0	50
<i>P. mirabilis</i>	33	43	29	57	0	-	-	15	14	14	0	100	57	57
<i>P. vulgaris</i>	100	33	0	100	0	-	-	0	0	33	0	33	33	67
<i>P. rustigianii</i>	0	100	0	0	0	-	-	100	0	0	0	100	0	0
<i>M. morgani</i>	100	100	50	75	12	-	-	12	0	0	12	37	11	0
<i>S. marcescens</i>	100	100	100	75	0	-	-	25	0	75	100	75	100	100
<i>P. aeruginosa</i>	95	95	86	93	85	46	59	5	33	52	40	90	79	90
<i>B. cepacia</i>	100	100	100	100	100	100	100	0	0	100	0	100	100	100
<i>P. putida</i>	100	100	100	100	100	100	100	0	100	100	100	0	100	100
<i>S. maltophilia</i>	100	100	100	100	100	0	100	100	25	75	100	100	50	25
<i>A. baumannii</i>	93	100	100	87	87	36	64	0	36	57	57	28	100	71
<i>F. odoratum</i>	100	100	100	100	0	0	0	0	0	0	0	0	0	0
<i>C. freundii</i>	100	100	50	100	0	100	0	0	0	0	0	100	100	0
<i>S. Infantis</i>	100	100	0	100	100	100	100	0	0	100	100	100	100	100
Non fermenter GNB	100	100	100	67	67	100	100	0	67	67	67	33	67	67
Total percentage	93	76	55	72	62	56	46	16	30	55	50	62	59	55

AMP- Ampicillin; CPL- Cephalotin; CFO- Cefoxitin; CRX- Cefuroxime; CRO- Ceftriaxone; CAZ- ceftazidime; CAR- carbenicillin; IMP- Imipenen; CIP- Ciprofloxacin; GEN- Gentamicin; AMI- Amikacin; TET- Tetracycline; CLO- Chloramphenicol; TSX- trimethoprim –Sulfametoxazole.

Table 6
General clinical-pathological data from newborns with diarrhoea caused by *Salmonella* serovar Infantis isolated from faeces in 1996

Patient	Date of isolation	Birth weight	Age (days)	Rupture of membranes	Prematurity	Type of partum	Cause of Admission
GG 1	05/07	1465 g	6	>24 hours	yes	Normal	RDS
GG 2	05/13	1105 g	37	<24 hours	yes	Cesarean	RDS
GG 3	05/07	2990 g	63	<24 hours	no	Normal	Asphyxia
GG 4	05/07	3470 g	8	-	no	Cesarean	Tachypneic
GG 5	05/07	3170 g	14	-	no	Cesarean	Gastric enteritis
GG 6	05/07	-	7	-	no	Normal	Jaundice + skin infection
GG 7	05/13	2810 g	7	<24 hours	yes	Normal	RDS
GG 8*	05/14	-	-	-	-	-	-
GG 9	05/20	1870 g	7	<24 hours	yes	Cesarean	RDS

RDS = Respiratory deficiency syndrome; *No available data.

resistance to ampicillin, cephalosporin (except cefoxitin), gentamicin and amikacin, with combinations of resistance to chloramphenicol, tetracycline and trimethoprim-sulfametoxazole. According to FRENCH *et al.* (1996), these strains of *Klebsiella sp.* do not respond to an empiric treatment with gentamicin or amikacin. LIVRELLI *et al.* (1996), emphasized that these bacterial strains are pathogens which cause infections, mainly on immuno-deficient patients.

36% of Methicillin-resistant *S. aureus* (MRSA) were found. This result matches the results of other places in Brazil and other countries (LEVY *et al.*, 1991; PANLILIO *et al.*, 1992; VOSS *et al.*, 1994; DURMAZ *et al.*, 1997; SADER *et al.*, 1999).

Despite of the relatively recent use of imipenem and ciprofloxacin in hospital, we observed a considerable level of resistance to gram

negatives. STRUELENS *et al.* (1993) verified that 40% of *A. baumannii* strains are resistant to the latter antimicrobial in Brussels.

The nosocomial outbreak caused by *Salmonella* serovar Infantis was the first one in the literature. It showed the potential virulence of a plasmid of high molecular weight, and the multiresistance of this serovar, similar to that of serovars Typhimurium and Agona in pathogenicity, virulence and potential to cause outbreaks of hospital infections.

Recent studies on salmonellosis show that Infantis and Agona serovars have been isolated in stools and blood of children in hospitals in Rio de Janeiro, with a significant prevalence of Typhimurium serovar (ASENSI, SOLARI & HOFER, 1994; ASENSI & HOFER, 1994). This *S. Infantis* outbreak probably resulted from the vulnerability of the neonates due to their severe primary processes which favored the infection of the bacterium. Among the risk factors (Table 6) were observed prematurity and respiratory problems at birth. Besides, all of the newborns were in a neonatal pathologic nursery, submitted to invasive procedures.

Another interesting aspect was the presence of the higher molecular weight plasmid (98Md), which was probably involved with codification of virulence genes in *Salmonella sp.* The presence of a 60Md plasmid of *S. Typhimurium* increases virulence in mouse models as compared with plasmid free or plasmid cured strains (JONES *et al.*, 1982; JONES & OSBORNE, 1991). For some years, various publications have illustrated the role of plasmids as virulence related factors in various serovars of *Salmonella*. Chromosomal DNA is an important factor in expression of virulence, especially in the capacity of strains to survive and multiply in reticuloendothelial system cells. Nevertheless, the virulence of *Salmonella* strains is linked to a combination of chromosomal and plasmid DNA, the last one related to adhesion and invasion of HeLa cells and mouse infection (JONES *et al.*, 1982; REXACH *et al.*, 1994). In 1985, HELMUTH *et al.* characterized several virulence-associated plasmids in different *Salmonella* serovars.

The Committee for Hospital Infection Control used the results of this study to control new cases, optimize the use of antimicrobial in the hospital, implement its policies to reduce excess hospital stay and costs of medical care.

RESUMO

Análise epidemiológica de cepas bacterianas envolvidas em infecção hospitalar em um Hospital Universitário no Brasil

As infecções hospitalares representam um aumento na morbidade e mortalidade de pacientes internados, com significativo aumento no custo de internação hospitalar. Teve-se como objetivo fazer uma análise epidemiológica de casos de infecção hospitalar ocorridos num Hospital Universitário na cidade do Rio de Janeiro. Assim, foram analisadas 238 cepas isoladas a partir de 14 espécimens clínicos diferentes oriundos de 166 pacientes internados no período de 08 de 1995 a 07 de 1997. A idade média dos pacientes foi de 33,4 anos, 72,9% faziam uso de antimicrobiano antes de apresentar a cultura positiva, as patologias de risco mais comuns foram: Cirurgia (19,3%), HIV ou AIDS positivo (18,1%) e Patologia Pulmonar (16,9%). Foram identificadas 24 espécies bacterianas distintas, com predominância de *S. aureus* (21%) e *P. aeruginosa* (18,5%). Foram detectados 36% de MRSA (Methicilin

Resistant *S. aureus*). Os Gram negativos apresentaram altos níveis de resistência para aminoglicosídeos e cefalosporinas. Foi detectado um surto de diarréia em berçário patológico, provocado pela *Salmonella* sorovar Infantis, com altos níveis de resistência para antimicrobianos e um plasmídeo de alto peso molecular (98Mda), codificador do fator R.

REFERENCES

1. AL-ORAINY, I.O.; AL-NASSER, M.N.; SAEED, E.S. & CHOWDHURY, M.N.H. - Nosocomial bacteraemia in a teaching hospital in Saudi Arabia. *J. Hosp. Infect.*, **14**: 201-207, 1989.
2. ASENSI, M.D. & HOFER, E. - Serovars and multiple drug resistant *Salmonella sp.* isolated from children in Rio de Janeiro-Brazil. *Rev. Microbiol. (S. Paulo)*, **25**: 149-153, 1994.
3. ASENSI, M.D.; SOLARI, C.A. & HOFER, E. - A *Salmonella agona* outbreak in a pediatric hospital in the city of Rio de Janeiro-Brazil. *Mem. Inst. Oswaldo Cruz*, **89**: 1-4, 1994.
4. AYLIFFE, G.A.J. - The progressive intercontinental spread of methicillin-resistant *Staphylococcus aureus*. *Clin. infect. Dis.*, **24** (suppl. 1): S74-S79, 1997.
5. BAIRD, G.D.; MANNING, E.J. & JONES, P.W. - Evidence for related virulence sequences in plasmids of *Salmonella dublin* and *Salmonella typhimurium*. *J. gen. Microbiol.*, **131**: 1815-1823, 1985.
6. BALOWS, A.; HAUSLER JR., W.J.; HERRMANN, K.L.; ISENBERG, H.D. & SHADOMY, H.J. - *Manual of clinical microbiology*. 5. ed. Washington, American Society for Microbiology, 1991.
7. BERGOGNE-BÉRÉZIN, E. - Les infections nosocomiales: nouveaux agents, incidence, prévention. *Presse méd.*, **24**: 89-97, 1995.
8. BIRNBOIM, H.C. & DOLY, J. - A rapid alkaline extraction procedure for screening recombinant plasmid DNA. *Nucleic Acids Res.*, **7**: 1513-1523, 1979.
9. COTTON, M.F.; BERKOWITZ, F.E.; BERKOWITZ, Z.; BECKER, P.J. & HENEY, C. - Nosocomial infections in black South African children. *Pediatr. infect. Dis. J.*, **8**: 676-683, 1989.
10. DUNCAN, J.M.G.; VALENCIA, E.; CELIZ, M.S.E. *et al.* - Vigilancia de la susceptibilidad in vitro de las bacterias intrahospitalarias. *Bol. Speit.*, **3**: 25-30, 1994.
11. DURMAZ, B.; DURMAZ, R. & SAHIN, K. - Methicillin-resistance among Turkish isolates the *Staphylococcus aureus* strains from nosocomial and community infections and their resistance patterns using various antimicrobial agents. *J. Hosp. Infect.*, **37**: 325-329, 1997.
12. FRENCH, G.L.; SHANNON, K.P. & SIMMONS, N. - Hospital outbreak of *Klebsiella pneumoniae* resistant to broad-spectrum cephalosporins and β -lactam- β -lactamase inhibitor combinations by hiperproduction of SHV-5 β -lactamase. *J. clin. Microbiol.*, **34**: 358-363, 1996.
13. HELMUTH, R.; STEPHAN, R.; BUNGE, C. *et al.* - Epidemiology of virulence-associated plasmids and outer membrane protein patterns within seven common *Salmonella* serotypes. *Infect. Immun.*, **48**: 175-182, 1985.
14. JARVIS, W.R. - Epidemiology of nosocomial infections in pediatric patients. *Pediatr. infect. Dis. J.*, **6**: 344-351, 1987.
15. JONES, C.S. & OSBORNE, D.J. - Identification of contemporary plasmid virulence genes in ancestral isolates of *Salmonella enteritidis* and *Salmonella typhimurium*. *FEMS Microbiol. Lett.*, **80**: 7-12, 1991.
16. JONES, G.W.; RABERT, D.K.; SVINARICH, D.M. & WHITFIELD, H.J. - Association of adhesive, invasive, and virulent phenotypes of *Salmonella typhimurium* with autonomous 60-Megadalton plasmids. *Infect. Immun.*, **38**: 476-486, 1982.

17. KOELEMAN, J.G.M.; PARLEVLIET, G.A.; DIJKSHOORN, L.; SAVELKOU, P.H.M. & VANDENBROUCKE-GRAULS, C.M.J.E. - Nosocomial outbreak of multi-resistant *Acinetobacter baumannii* on a surgical ward: epidemiology and risk factors for acquisition. **J. Hosp. Infect.**, 37: 113-123, 1997.
18. LEVY, C.E.; MONTELLI, A.C.; FURTADO, J.S. *et al.* - Resistência a drogas em cepas bacterianas de serviços hospitalares: Laboratório de referência do sistema COBA. **Rev. Microbiol.** (S. Paulo), 22: 21-27, 1991.
19. LIVRELLI, V.; CHAMPS, C.; MARTINO, P. *et al.* - Adhesive properties and antibiotic resistance of *Klebsiella*, *Enterobacter* and *Serratia* clinical isolates involved in nosocomial infections. **J. clin. Microbiol.**, 34: 1963-1969, 1996.
20. MACRINA, F.L.; KOPECKO, D.J.; JONES, K.R.; MAYERS, D.J. & McCOWAN, S.M. - A multiple plasmid-containing *Escherichia coli* strain: convenient source of size reference plasmid molecules. **Plasmid**, 1: 417- 420, 1978.
21. MANRIQUE, E.I. & GALVÃO, L.L. - Racionalização e controle de antimicrobianos. In: RODRIGUES, E.A.C.; MENDONÇA, J.S. de; AMARANTE, J. *et al.* - **Infeções hospitalares: prevenção e controle**. São Paulo, Sarvier, 1996.
22. MINISTRY OF HEALTH - BRAZIL / Ministério da Saúde-Brasil. Vigilância Epidemiológica por Componentes - Original Title: National Nosocomial Infection Surveillance System (NNISS-USA), 1994. 102 p.
23. MONNET, D.L.; BIDDLE, J.W.; EDWARDS, J.R. *et al.* - Evidence of interhospital transmission of extended-spectrum β -Lactam-resistant *Klebsiella pneumoniae* in the United States, 1986 to 1993. **Infect. Control Hosp. Epidem.**, 18: 492-498, 1997.
24. NATIONAL COMMITTEE FOR CLINICAL LABORATORY STANDARDS - Performance standards for antimicrobial disk susceptibility tests. Approved Standard. NCCLS Publication M2-A5., 1993.
25. PANLILIO, A.L.; CULVER, D.G. & GAYNES, R.P. - Methicillin-resistant *Staphylococcus aureus* in US hospitals, 1975-1991. **Infect. Control Hosp. Epidem.**, 13: 582-586, 1992.
26. REXACH, L.; DILASSER, F. & FACH, P. - Polymerase chain reaction for *Salmonella* virulence-associated plasmid genes detection: a new tool in *Salmonella* epidemiology. **Epidem. Infect.**, 112: 33-43, 1994.
27. SADER, H.S.; SAMPAIO, J.L.M.; ZOCCOLI, C. & JONES, R.N. - Results of the 1997 SENTRY Antimicrobial Surveillance Program in three Brazilian medical centers. **Braz. J. infect. Dis.**, 3: 63-79, 1999.
28. SAMBROOK, J.; FRITSCH, E.F. & MANIATS, T. - Molecular cloning: a laboratory manual. 2. ed. New York, Cold Spring Harbor Laboratory, 1989. p. 1-28.
29. SARTOR, C.; SAMBUC, R.; BIMAR, M.C.; GULIAN, C. & DE MICCO, P. - Prevalence surveys of nosocomial infections using a random sampling method in Marseille hospitals. **J. Hosp. Infect.**, 29: 209-216, 1995.
30. SMITH, S.D. & DOEBBELING, B.N. - Costs of nosocomial infections. **Curr. Opin. infect. Dis.**, 9: 286-290, 1996.
31. STROUD, L.; SRIVASTAVA, P.; CULVER, D. *et al.* - Nosocomial infection in HIV-infected patients: preliminary results from a multicenter surveillance system (1989-1995). **Infect. Control Hosp. Epidem.**, 18: 479-485, 1997.
32. STRUELENS, M.J.; CARLIER, E.; MAES, N. *et al.* - Nosocomial colonization and infection with multiresistant *Acinetobacter baumannii* outbreak delineation using DNA macrorestriction analysis and PCR-fingerprinting. **J. Hosp. Infect.**, 25: 15-32, 1993.
33. THRELFALL, E.J.; ROWE, B.; FERGUSON, J.L. & WARD, L.R. - Characterization of plasmids conferring resistance to gentamicin and apramycin in strains of *Salmonella typhimurium* phage type 204c isolated in Britain. **J. Hyg. (Lond.)**, 97: 419-426, 1986.
34. VOSS, A.; MILATOVIĆ, D.; WALLRAUCH-SCHWARZ, C.; ROSDALL, V.T. & BRAVENEY, I. - Methicillin-resistant *Staphylococcus aureus* in Europe. **Europ. J. clin. Microbiol. infect. Dis.**, 13: 50-55, 1994.
35. WENZEL, R.P. - Epidemiology of hospital-acquired infection. In: BALLOWS, A., ed. **Manual of clinical microbiology**. 5. ed. Washington, American Society for Microbiology, 1994. p. 147-150.

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