

RESEARCH ON BACTERIAL RESISTANCE TO ANTIBIOTICS, IN SUSPICION OF NOSOCOMIAL INFECTIONS, IN MAJOR HOSPITALS IN BUCHAREST

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Abstract

According to the Romanian Ministry of Health, nosocomial infection (NI) is the infection acquired in hospitals or other health care facilities with beds and refers to "any disease which can be clinically or microbiologically recognized affecting either the patient, because of his admission to hospital or care received as a patient or outpatient hospital or the medical staff due to its activity, whether or not symptoms occur while the person is in hospital."

In our research, for some of the bacteria present in the selected samples group suspected of there were performed susceptibility/resistance tests to frequently used antibiotics. The germs in question were mainly resistant to antibiotics. The results support the suspected presence of NI, known to be due to germs from a multi-resistant hospital flora. Susceptibility was not good at any of the tested antibiotics, as expected for NI bacteria, which are often multi-drug resistant.

We observed that the phenomenon of nosocomial infections exists, it is not frequent, but usually not reported. In the present study, from the total number of 3,248 completed antibiograms.

Rezumat

Conform Ministerului Sănătății, infecția nosocomială (NI) reprezintă „infecția contractată în spital sau alte unități sanitare cu paturi” și se referă la „orice boală ce poate fi recunoscută clinic sau microbiologic, care afectează fie bolnavul, datorită internării lui în spital sau îngrijirilor primite, ca pacient spitalizat sau în tratament ambulatoriu, fie personalul sanitar, datorită activității sale, indiferent dacă simptomele bolii apar sau nu în timp ce persoana respectivă se află în spital”.

În cercetarea noastră, pentru o parte din bacteriile prezente în grupul de prelevate selectat, cu suspiciune de N.I., au fost efectuate teste de sensibilitate/rezistență la antibiotice de uz curent. S-a observat că la germenii incriminați predomină rezistența la antibiotice. Rezultatele conduc la susținerea ideii de suspectare a prezenței unor NI, despre care se știe că sunt cauzate de germeni proveniți dintr-o floră spitalicească multirezistentă. Sensibilitatea nu a fost bună la niciunul din antibioticele testate, așa cum era de așteptat pentru bacteriile din N.I., care adesea sunt multirezistente.

S-a observat că fenomenul N.I. există, fără a fi frecvent, dar nu se raportează de obicei. În studiul prezent dintr-un total de 3.248 antibiograme realizate, 17 sunt pentru N.I., dintre care numai 11 sunt cu diagnostic clinic, care orientează spre N.I., conform biletului de trimitere la laborator și numai 8 sunt confirmate microbiologic.

Secrețiile purulente de plagă trimise pentru analiză la laborator de la secțiile chirurgicale pot fi asimilate cu infecțiile nosocomiale, care sunt rareori raportate în spitalele din România.

Keywords: antibiotics, bacterial resistance, NI (nosocomial infection), antibiogram, hospital flora, antibiotics

Introduction

The study of bacterial resistance in suspected nosocomial infections (NI) is a serious problem in the anti-infective therapy and is in the attention of the World Health Organization (WHO) and the European Union (EU) [19, 20, 21, 23]. In the present work, a statistical retrospective / prospective study has been undertaken on sensitivity and resistance to antibiotics of pathogenic bacteria isolated in a Romanian university hospital, taking the antibiogram data from clinical microbiology and sanitary microbiology laboratories [13, 21].

Our objective was to study this phenomenon and to make a retrospective and prospective research of bacterial sensitivity and resistance, in custom conditions of Bucharest, Romania, which can be considered as representative for the population of this country.

NI term is defined differently, covering various aspects of infections acquired within hospitalisation [6, 7, 11, 14]. In Romania, the Ministry of Health (MH) regulations governing supervision and control of NI, define it as "infection acquired in hospitals or other health care facilities with beds" and refers to "any disease that can be recognized clinically or microbiologically, affecting either patient due to his admission to hospital or care received either as a patient or outpatient hospital or medical staff, because of its activity, whether or not symptoms occur while the person is in hospital."

The MH Order no. 994/2004 defines 13 major categories of NI [24]. For these categories, in this study there were considered a number of infections that presented specific events for NI [5, 9, 12, 15, 16, 17, 18]. Thus, for surgical infections we discuss mainly about wound infections, surgical incision or deep surgical wound infections, which may develop particular issues [1, 2, 3, 10].

Materials and Methods

The samples considered for the statistical study of microbiological results comprised 3248 antibiograms performed in clinical and sanitary microbiology laboratories within a clinical hospital in Bucharest.

The analysis of samples conforms to the WHO regulations, MH and the Romanian Academy of Medical Sciences (RASM), namely the disk diffusion susceptibility test, after Kirby and Bauer method [4, 8]. Antibiotic

susceptibility tests were performed on Mueller-Hinton agar seeded with microbial suspension of microbial strain culture from patients in Ø 10 cm Petri dishes, aerobically incubated for 24 hours. The reading was performed by measuring the inhibition zone diameter in millimeters and reporting to the table interpretation in SIR system (Sensitive, Intermediate and Resistant).

Results and Discussion

Reagent-test antibiotics used were standardized (Labormed, Oxoid biodisks, Difco sensidisks) and not-standardized antibiogram microdisks. Of the total number of tests, most have been standardized, namely a number of 35,208.

Bacterial susceptibility testing was carried out against 55 antibacterial chemotherapy, 25 of them being commonly used in medical practice. (Table I)

Table I
Frequency of standardized antimicrobial chemotherapy used in testing, sorted by rank

No.	Antibacterial chemotherapy	Test no.	%	Rank
1.	Gentamicin	2530	7.19	1
2.	Colistin	2393	6.80	2
3.	Kanamycin	2328	6.61	3
4.	Co-trimoxazole	2307	6.55	4
5.	Rifampicin	2228	6.33	5
6.	Chloramphenicol	2152	6.11	6
7.	Oxacillin	2111	6.00	7
8.	Streptomycin	2095	5.95	8
9.	Tetracycline	2063	5.86	9
10.	Erythromycin	1961	5.57	10
11.	Augmentin	1580	4.49	11
12.	Ampicillin	1502	4.27	12
13.	Penicillin	1479	4.20	13
14.	Amoxicillin	1442	4.10	14
15.	Neomycin	1254	3.56	15
16.	Cephalothin	1235	3.51	16
17.	Lincomycin	1012	2.87	17
18.	Nalidixic acid	892	2.53	18
19.	Nitrofurantoin	770	2.19	19
20.	Doxycyclin	632	1.80	20
21.	Novobiocin	588	1.67	21
22.	Sulfafurazol	274	0.78	22
23.	Furazolidone	204	0.58	23
24.	Carbenicillin	122	0.35	24
25.	Polymyxin	54	0.15	25
TOTAL		35,208		

Note: Rankings are descending, in the order of frequency, in absolute terms.

A number of 14 bacterial species were isolated and identified, and a total of 1,363 tested strains.(Table II).

Table II
Frequency and rank of isolated bacteria, identified and tested by
antibiogram in microbiology laboratories within major hospitals in
Bucharest

No.	Identified germ	Tested strains no.	%	Rank
1.	<i>Escherichia coli</i>	674	49.45	1
2.	<i>Staphylococcus aureus</i>	361	26.49	2
3.	<i>Enterobacter</i> sp.	95	6.97	3
4.	<i>Proteus</i> sp.	61	4.48	4
5.	<i>Klebsiella</i> sp.	48	3.52	5
6.	<i>Staphylococcus saprophyticus</i>	40	2.93	6
7.	<i>Bacillus cereus</i>	19	1.39	7
8.	<i>Pseudomonas aeruginosa</i>	16	1.17	8
9.	Beta-hemolytic streptococcus	15	1.10	9
10.	<i>Citrobacter</i> sp.	12	0.88	10
11.	<i>Providentia</i> sp.	10	0.73	11
12.	<i>Streptococcus pyogenes</i> grup A	6	0.44	12
13.	<i>Streptococcus viridans</i>	4	0.29	13
14.	<i>Streptococcus</i> sp.	2	0.15	14
	TOTAL	1,363	100.00	

In terms of recommendations for the antibiogram, it was noticed that some of the isolated germs were not identified and tested in the laboratory, but the antibiogram was still performed. Under these conditions, for not respecting the recommendations for sensitivity/resistance testing, data stored for these germs were not statistically processed.

Statistical processing of data on **antibacterial chemotherapy** used for susceptibility testing revealed the use of a wide variety of groups of antibiotics and antibacterial chemotherapy, which covers almost all the therapeutic antibacterial agents used in Romania. It is noticeable the variable number of tests processed statistically, some substances being frequently used, others very rarely.

The antibiograms were performed on germs isolated and tested in the laboratory, even if some were not identified. Each strain was tested at several antibacterial chemotherapy, on average 10.8 were used for testing antibiotics/antibiogram. The number of isolated, identified and tested species / bacterial groups was 14, out of 28 entities isolated and tested by antibiogram (ABG), some of them are identified as species and others as a group or by certain biochemical characteristics, especially for “hospital flora” in the environment [5, 9]. It is to be mentioned that within the

international references, "Hospital Pharmacist Report 1996" USA [22], the antibiogram identified 71 bacterial species and groups, that were tested on 62 antibiotics. Meanwhile, for the period between 2001-2008, the Surveillance System for Antimicrobial Resistance in Europe presented the EARSS 2009 report on 6 multidrug-resistant bacterial species and five groups of antibiotics [21].

Analysis for clinical microbiology were made mostly for medical ward, ophthalmology, surgery, dermato-venereal, ENT, urology and ambulatory, and sanitary microbiology tests were performed on "hospital flora", for regular monitoring of medical staff risk and for NI.

In the study, microbiological determinations were performed on 13 types of samples contaminated with pathogenic bacteria, totaling 3248 samples, of which the majority were eye discharge, urine culture and nasal exudate. Purulent secretions of plague sent to the laboratory for analysis from surgical wards can be clinically assimilated with NI, which are rarely reported in Romanian hospitals. (Table III)

Table III

Frequency and rank of samples of which isolated strains were tested by antibiogram in the microbiology laboratories within major hospitals in Bucharest

No.	Analyzed microbiological samples	Sample no.	%	Rank	Observations
1.	Eye discharge	952	29.31	1	very frequently
2.	Urine culture	857	26.39	2	very frequently
3.	Nasal exudate	703	21.64	3	very frequently
4.	Pharyngeal exudate	557	17.15	4	very frequently
5.	Ear discharge	61	1.88	5	frequently
6.	Wound secretion	28	0.86	6	frequently
7.	Vaginal discharge	19	0.58	7	frequently
8.	Pus boil discharge	18	0.55	8	frequently
9.	Sputum	16	0.49	9	frequently
10.	Stool	13	0.40	10	frequently
11.	Urethral discharge	10	0.31	11	frequently
12.	Conjunctival discharge	4	0.12	13	rarely
13.	Gall	4	0.12	13	rarely
14.	<i>Not specified</i>	6	0.18	12	incorrect*
	TOTAL	3,248			

* - In this case is not recommended to perform antibiogram.

Within the analyzed period, with data collected at random from 2000-2010, we identified 17 probable cases of NI, but the hospital studied did not report explicitly any of them.

The attitude of underestimating the frequency of NI is prevalent everywhere, but in Romania there are hospitals reporting zero NI in statistical situations. If the phenomenon is minimalized, the causes will persist, aggravating the patient's condition, increasing bacterial resistance to antibiotics and rising social and financial costs of the treatment (Table IV).

Table IV
Nosocomial infections suspected

No.	Sample	Pathogens	Observations
1.	Wound secretion	<i>E. coli</i>	Isolated and identified
2.	Wound secretion	<i>E. coli</i>	Isolated and identified
3.	Wound secretion	Haemolytic flora GP*	Isolated
4.	Ankle wound secretion	<i>Pseudomonas aeruginosa</i>	Isolated and identified
5.	Wound discharge pus	Haemolytic streptococcus	Isolated and identified
6.	Pus discharge	Haemolytic flora GP	Isolated
7.	Pus discharge	Haemolytic flora GP	Isolated
8.	Tube discharge	<i>E. coli</i>	Isolated and identified

*GP – gram-positive

For some of the bacteria present in selected samples group with suspected NI there were performed sensitivity / resistance tests to commonly used antibiotics. It was noticed that incriminated germs are more than twice as resistant: sensitivity 18.70% to resistance 40.43% (figure 1). One can appreciate that the results support the idea of NI suspicion, which are known to be mainly caused by germs coming from multi-resistant hospital flora. Sensitivity was not good at any of the antibiotics tested, as expected for NI bacteria, which are often multi-drug resistant. Resistance was recorded for antibiotics Rifampicin, Gentamicin, Furazolidone (over 80%).

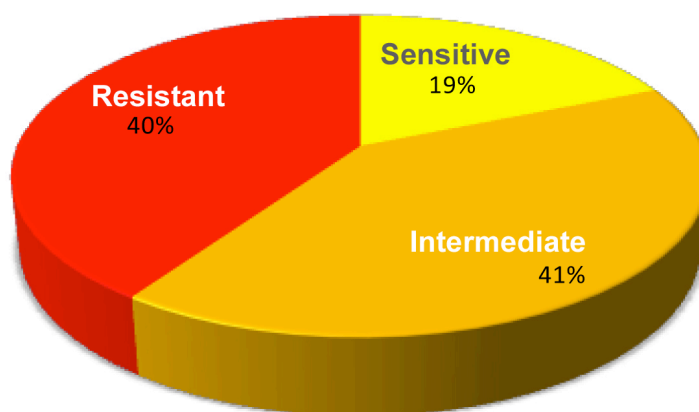


Figure 1.

Bacterial spectrum of sensitivity/resistance to antibiotics in nosocomial infections

The tested bacteria were gram-negative bacilli and gram-positive cocci, but neither species was not sufficient for statistical processing. It was compared the sensitivity of isolated and identified bacteria from pus taken from different infections. Another six "purulent discharge" were investigated during our study, but we did not include these in the statistical processing because the antibiogram was performed on isolated, but unidentified flora. To these, only a few results were "sensitive", 22 out of 122 tests (approximately 18%), suggesting that it is a multi-resistant hospital flora, so perhaps these cases are also NI. Overall, the sensitivity of isolated and identified bacteria from "wound pus" is very low compared to the ear and eye pus, so we are probably in the presence of germs coming from multiresistant hospital flora.

We observed that the phenomenon of NI is not frequent, but usually not reported. From a total of 3248 susceptibility tests, 17 are for NI, of which only 11 are as clinical diagnosis, according to laboratory reference ticket and 8 are microbiologically confirmed. We mention that in the clinical hospital in Bucharest where we conducted research, in the study period, there were no serious cases of NI, as they were in recent years publicized cases in Romania.

Conclusions

The statistical processing of antibiogram results, filtered by samples, bacteria and antibiotics reveals the suspicion of NI during the investigated period. The phenomenon of nosocomial infections is not frequent, but usually not reported; generally, hospitals report partially or sometimes do not report cases of NI. From a total of 3248 antibiograms, only 17 are for nosocomial infections susceptibility, of which only 11 are certain, according to the laboratory reference ticket. Bacterial strains isolated and identified in samples with suspected NI are multidrug-resistant, the highest resistance (over 80%) was recorded for Rifampicin, Gentamicin, Furazolidone. Statistical processing of the other data acquired and stored in the database we created will be useful for understanding the dimension of bacterial sensitivity and resistance to antibiotics.

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