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RISK FACTORS FOR EXTENDED-SPECTRUM β-LACTAMASE-PRODUCING *ESCHERICHIA COLI* INFECTION IN HOSPITALIZED PATIENTS

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ABSTRACT

The incidence of nosocomial infection caused by extended-spectrum β-lactamase (ESBL)-producing bacteria is increasing worldwide. Infections caused by ESBL producers have been associated with severe adverse clinical outcomes that have led to increased mortality, prolonged hospitalization, and rising medical costs. To avoid such adverse events and ineffective treatment, an appropriate use of drugs for infectious diseases is needed. To suppress the emergence and spread of drug-resistant bacteria in hospitals, it is important to be vigilant about ESBL-producing *Escherichia coli* (*E. coli*). In this study, we examined and compared seven items in a blood test between patients with ESBL-producing *E. coli* and non-ESBL-producing *E. coli* among febrile patients. We examined the levels of serum albumin, hemoglobin, and C-reactive protein (CRP), and the numbers of leucocytes, neutrophils, lymphocytes, and platelets in blood on the day of admission, the screening day during hospitalization, and the day immediately before discharge from the hospital. There were no significant differences in clinical background characteristics between the two groups of patients. In patients with invasive infections caused by ESBL-producing *E. coli*, serum albumin levels and the number of lymphocytes were significantly lower than those in patients not infected with ESBL producers. These values recovered to their baseline levels on the day of hospital discharge. This retrospective study suggests that serum albumin levels and the number of lymphocytes may serve as risk factors for infection by ESBL-producing *E. coli*, thereby supporting the appropriate use of antimicrobials in hospitals.

Key Words: Extended-spectrum β-lactamase, ESBL, *Escherichia coli*, Risk factor, Drug-resistant bacterium

INTRODUCTION

The emergence of nosocomial extended-spectrum β-lactamase (ESBL)-producing *Klebsiella pneumoniae* and *Serratia marcescens* was first reported approximately 30 years ago.[1)] The incidence of infection by ESBL producers has been increasing not only in hospitals, but also in communities.[2-4)] In the USA and Europe, the excessive use of inexpensive drugs, such as

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cephalosporins, will likely lead to the widespread emergence of ESBL producers.[5,6] In Japan, such producers have been increasing from the latter half of the 1990s.[7] Infections by ESBL producers have been associated with severe adverse clinical outcomes that have led to increased mortality, prolonged hospitalization, and rising medical costs.[8] Those adverse outcomes have also been related, at least in part, to a delay in the administration of an effective therapy.[9-12] To avoid such adverse events, the appropriate use of drugs for infectious diseases will be needed.

There have been many reports on the risk factors for nosocomial infections, such as the recent use of antimicrobials and indwelling catheters (urinary catheters and tracheal tubes).[13-17] However, it is sometimes impossible when performing surgeries to limit catheter use in certain therapies. In addition, such risk factors are common to infectious diseases other than those caused by ESBL producers. Therefore, it is difficult to prevent infections by ESBL producers.

Because ESBL-producing *Escherichia coli (E. coli)* is the most common problem in our hospitals among the many bacterial species of ESBL producers mentioned above, in this study we focused on this microorganism. To suppress the emergence and spread of drug-resistant bacteria in our hospital, it was very important to be vigilant about controlling ESBL-producing *E. coli*.

Thus, we surveyed and compared seven items in a blood test conducted in patients infected and not infected with ESBL-producing *E. coli* among our febrile patients.

METHODS

Study design, population, and definition:

Data were collected between January 2009 and December 2010 in the Japanese Red Cross Nagoya Daiichi Hospital. Febrile patients were subjected to a bacterial culture test followed by ESBL screening tests. In the screening test, the bacteria from 69 patients proved resistant to ≥2 μg/ mL of ceftadizime, cefotaxime, or aztreonam. Consequently, we judged those to be ESBL-producing *E. coli* when we detected more than a 5-mm diameter zone between a ceftadizime alone-treated disk and a ceftadizime/clavulanic acid-treated disk. Total of 41 patients were excluded from this study because they were less than 18 years old and/or infected in the community, not in this hospital. Therefore, 14 patients infected with ESBL-producing *E. coli* and 14 age- and gender-matched patients not infected with ESBL-producing *E. coli* that were identified by ESBL confirmatory tests, were studied. Bacterial infection was determined by a broth microdilution method in accordance with the Clinical and Laboratory Standards Institute (CLSI) guidelines.[18] Our retrospective study was approved by the Institutional Review Board of the Japanese Red Cross Nagoya Daiichi Hospital (Approval No. 98).

To compare the clinical background characteristics of inpatients, we determined their age, gender, and body mass index (BMI); comorbidities; the hospital ward to which they were admitted; any history of hospitalization for the past three months; invasive devices used (urethral catheter, tracheal tube, central venous catheter and others); infected regions [urinary tract (urine samples), respiratory tract (respiratory samples), blood (blood samples), intestinal tract (stool samples), abdominal drains and wounds (other samples)]; number of days until detection of an ESBL producer during hospitalization; and a history of infection by *Pseudomonas aeruginosa*, methicillin-resistant *Staphylococcus aureus* (MRSA), or Candida. We also determined the number of patients who had used antimicrobials (classified according to the type of antimicrobial used), the total number of antimicrobials used, the total number of days of antimicrobial use, the maximum number of days of use of the same antimicrobial, and the number of patients treated with two or more antimicrobials. We also examined the levels of serum albumin, hemoglobin, and C-reactive protein (CRP) and the numbers of leucocytes, neutrophils, lymphocytes, and

platelets in blood on the day of admission, the screening days during hospitalization (at least once a week), and immediately before discharge.

Data analyses:

Data were analyzed by the Wilcoxon signed-rank test (Fig. 1) and the Mann-Whitney U-test or Fisher's exact test (Tables 1 and 2), respectively. The results were expressed for each group

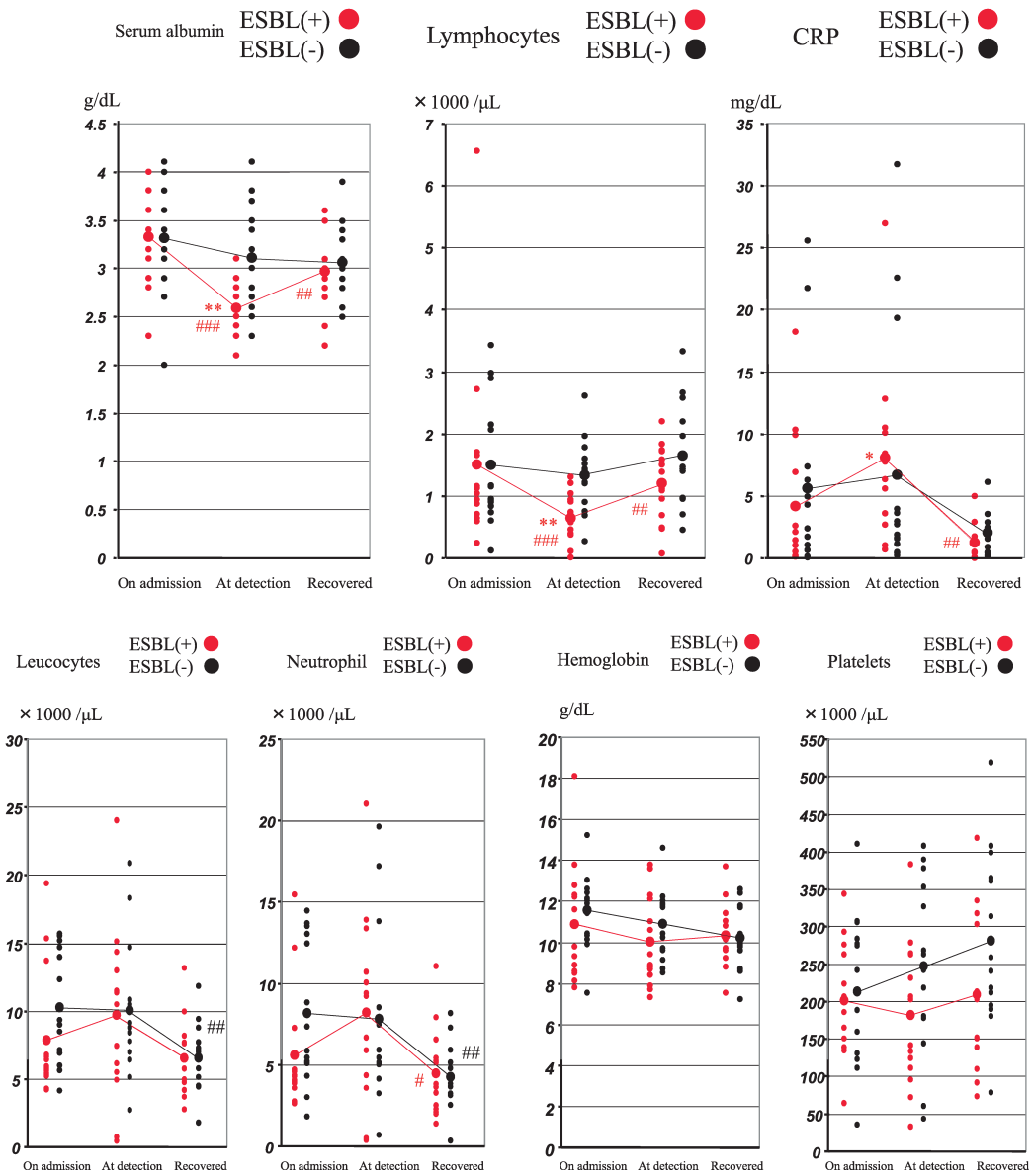

Fig. 1 Changes in biochemical parameters in blood on the day of admission (On admission), during hospitalization (At detection), and after hospitalization (Recovered). ESBL (+): patients infected with ESBL-producing *E. coli*. ESBL (–): patients not infected with ESBL-producing *E. coli*. **P*<0.05, ***P*<0.01 vs. On admission, $^{\#}P$<0.05, $^{\#\#}P$<0.01 vs. At detection, $^{\#\#\#}P$<0.05 vs. ESBL (–).

Table 1 Clinical background characteristics of patients infected and not infected with extended-spectrum β-lactamase (ESBL)-producing *E. coli*

Variable	ESBL (+) n=14 (%)	ESBL (–) n=14 (%)	*P*
Demographics			
Age (year): mean ± SD	72.1 ± 11.1	67.2 ± 16.7	*N.S.
Gender: Number of males	8 (57.1)	8 (57.1)	N.S.
BMI (mean ± SD)	20.0 ± 3.7	19.9 ± 3.6	*N.S.
Comorbidities			
Respiratory disease	3 (21.4)	3 (21.4)	N.S.
Cardiovascular disease	3 (21.4)	2 (14.3)	N.S.
Hypertension	6 (42.9)	7 (50.0)	N.S.
Diabetes mellitus	5 (35.7)	6 (42.9)	N.S.
Renal disease	3 (21.4)	2 (14.3)	N.S.
Liver disease	0 (0)	0 (0)	N.S.
Malignant disease	9 (64.3)	8 (57.1)	N.S.
Use of corticosteroids	2 (14.3)	4 (28.6)	N.S.
Hospital admission ward			
ICU	3 (21.4)	3 (21.4)	N.S.
Medical	8 (57.1)	6 (42.9)	N.S.
Surgical	3 (21.4)	5 (35.7)	N.S.
Previous hospitalization within 3 months	3 (21.4)	5 (35.7)	N.S.
Type of invasive device			
Urethral catheter	4 (28.6)	5 (35.7)	N.S.
Tracheal tube	2 (14.3)	2 (14.3)	N.S.
Central venous catheter	8 (57.1)	5 (35.7)	N.S.
Other	2 (14.3)	3 (21.4)	N.S.
Infected regions			
Urine samples	6 (42.9)	5 (35.7)	N.S.
Respiratory samples	2 (14.3)	1 (7.1)	N.S.
Blood samples	1 (7.1)	2 (14.3)	N.S.
Stool samples	2 (14.3)	0 (0)	N.S.
Other samples	3 (21.4)	5 (35.7)	N.S.
Days until detection of ESBL producer during hospitalization (mean ± SD)	36.4 ± 35.3	21.7 ± 12.5	*N.S.
Previous isolates			
Pseudomonas aeruginosa	3 (21.4)	3 (21.4)	N.S.
MRSA	2 (14.3)	1 (7.1)	N.S.
Candida	2 (14.3)	2 (14.3)	N.S.

ESBL (+): patients infected with ESBL-producing *E. coli*; ESBL (–): patients not infected with ESBL-producing *E. coli*; N.S.: not significant; BMI: body mass index; ICU: intensive care unit; MRSA: methicillin-resistant *Staphylococcus aureus*. Renal disease was defined as having a serum creatinine level >2 mg/dL. All statistical tests were performed using Fisher's exact test except for data indicated by *, for which Mann-Whitney U-test was used.

Table 2 Antimicrobials used for patients infected and not infected with extended-spectrum β-lactamase (ESBL)-producing *E. coli*

	ESBL (+) n=14 (%)	ESBL (-) n=14 (%)	*P*
Penicillins	4 (28.6)	3 (21.4)	N.S.
1st-generation cephalosporins	6 (42.9)	4 (28.6)	N.S.
2nd-generation cephalosporins	3 (21.4)	3 (21.4)	N.S.
3rd-generation cephalosporins	2 (14.3)	3 (21.4)	N.S.
4th-generation cephalosporins	8 (57.1)	4 (28.6)	N.S.
1st-4th generation cephalosporins	10 (71.5)	10 (71.5)	N.S.
Lyncosamide	2 (14.3)	1 (7.1)	N.S.
Carbapenems	2 (14.3)	1 (7.1)	N.S.
Tetracycline	1 (7.1)	0 (0)	N.S.
Aminoglycosides	2 (14.3)	0 (0)	N.S.
Fluoroquinolones	1 (7.1)	1 (7.1)	N.S.
Anti-MRSA	1 (7.1)	0 (0)	N.S.
Number of antimicrobials used	12 (85.7)	12 (85.7)	N.S.
Days of antimicrobial use (mean ± SD)	12.8 ± 13.5	7.6 ± 4.7	*N.S.
Number of patients treated with two or more antimicrobials	3 (21.4)	1 (7.1)	N.S.
Maximum number of days of use of same antimicrobial	10.1 ± 9.9	6.1 ± 3.5	*N.S.

ESBL (+): patients infected with ESBL-producing *E. coli*; ESBL (–): patients not infected with ESBL-producing *E. coli*; N.S.: not significant. All statistical tests were performed using Fisher's exact test except for data indicated by *, for which Mann-Whitney U-test was used.

as means ± SD in Tables 1 and 2. $P<0.05$ indicated statistical significance, when detected, a multivariate logistic regression analysis was conducted and the odds ratio was estimated.

RESULTS

There were no significant differences found between the two groups in terms of the clinical background characteristics of the patients (Table 1). On the day of admission to the hospital and during hospitalization, there were no significant differences between the two groups in terms of the number of patients who used antimicrobials (classified according to the type of antimicrobial used), the total number of antimicrobials used, the total number of days of antimicrobial use, the maximum number of days of use of the same antimicrobial, and the number of patients treated with two or more antimicrobials (Table 2).

On the day of admission, we could not detect any differences in the levels of serum albumin, hemoglobin, and CRP, or in the numbers of leucocytes, neutrophils, lymphocytes, and platelets in blood between the two groups. When infections were detected, serum albumin levels and the number of lymphocytes were significantly lower in patients infected with ESBL-producing *E.*

coli than those in patients not with ESBL-producing *E. coli*. The leucocyte curve and CRP curve of patients infected with ESBL producers were similar, whereas those in patients not infected with ESBL-producing *E. coli* were different (Fig. 1). Hemoglobin and platelet levels were unaffected throughout this study. In both groups, neutrophils reached normal levels on the day of discharge from the hospital, and no significant differences were observed between them during hospitalization. In many cases, owing to appropriate antimicrobial use (particularly carbapenems and aminoglycosides), the values returned to their baseline levels on the day of discharge from the hospital (Fig. 1). When we detected significant differences in the serum albumin levels and the number of lymphocytes between the two groups, a multivariate logistic regression analysis was conducted and the odds ratio was estimated. The adjusted odds ratios (95% confidence interval) of the serum albumin level (<3.0 g/dL) and the number of lymphocytes (<1,000 /μL) were 16.6 (1.4–205.0) (P=0.028) and 8.8 (1.2–65.6) (P=0.034), respectively, suggesting that each factor was significant for ESBL infection.

DISCUSSION

Our Japanese Red Cross Nagoya Daiichi Hospital serves as a main hospital in Nagoya. It consists of 34 diagnosis departments and 852 beds, and is typical of any large general hospital in Japan. In our hospital as well as hospitals worldwide, there is an increasing incidence of ESBL-producing *E. coli* infections that have led to severely adverse clinical outcomes. Some reasons have been cited for the nosocomial increase of this bacterium: 1) disuse of efficient/potent carbapenems; 2) increased use of 3rd-generation cephalosporins and quinolones in the community;[19] 3) stool-mediated infections;[20] and 4) existence of the ESBL gene in the plasmid.[2] In this study, we could not detect any statistical correlation between ESBL-producing *E. coli* infection and the number of patients who used antimicrobials (classified according to type of antimicrobial used), the total number of antimicrobials used, the total number of days of antimicrobial use, the maximum number of days of use of the same antimicrobial, and the number of patients treated with two or more antimicrobials.

The Centers for Disease Control and Prevention (CDC) in the USA has released guidelines for the prevention of multidrug-resistant gram-negative bacillus infection.[21] The guidelines propose general standard precautions for the infection at all medical facilities, particularly infection by multidrug-resistant organisms (MDROs), including ESBL-producing bacteria. The guidelines recommend comprehensive strategies, including 1) administrative support, 2) education of medical staff, 3) judicious use of antimicrobial agents, 4) MDRO surveillance, 5) infection control precautions, 6) frequent washing and disinfection of medical devices, and 7) decolonization. The guidelines for European hospitals have recommended that stool samples from all patients be examined on the day of admission to screen for ESBL-producing bacteria.[22] According to those guidelines, we performed an examination on admission to our hospital.

It is necessary to screen patients because the number of those infected with ESBL-producing bacteria has seen an increase recently in the community. Therefore, on admitting a patient who is suspected of infection and referred to us by other hospitals, a culture of the patient's sputum or urine samples was performed to confirm whether or not the patient is infected with ESBL-producing bacteria. In addition to the CDC guidelines, the medical staff and inpatients were asked to frequently gargle and wash their hands, and a private room was assigned to the patients infected with ESBL-producing bacteria.

In our hospital, we conducted blood tests in infectious patients at least once a week to confirm any effects of the treatment during hospitalization. Generally, the serum albumin level was used to

evaluate nutrition status. In addition, each item in the blood test served as an index: hemoglobin was the index of anemia; CRP, of acute inflammation; leucocytes, of infection and inflammation; neutrophils, of bacterial infection; lymphocytes, of viral infection; and platelets, of hemostasis. In this study, serum albumin levels and the number of lymphocytes were significantly lower in patients infected with ESBL-producing *E. coli* than in patients without it.

Reduction of serum albumin levels was a sign of malnutrition and induced the increase of mortality and morbidity rates.[23,24] Hypoalbuminemia was the result of the combined effects of inflammation and inadequate protein and calorie intake in patients with chronic diseases, such as chronic renal failure.[25] Furthermore, inflammatory cytokines, including interleukin-1β (IL-1β), interleukin-6 (IL-6), and tumor necrosis factor-α (TNF-α), have been reported to inhibit the synthesis of albumin.[26] However, we could not find any significant correlation between those clinical backgrounds and the low serum albumin levels before ESBL-producing *E. coli* infection. The time interval between "on admission" and "at detection" was less than 10 days, and generally the half-life of albumin was approximately 3 weeks,[27,28] suggesting that the low serum albumin levels may not be caused by ESBL-producing *E. coli* infection. Although the exact cause was not clear, on the day the ESBL-producing *E. coli* infection was detected, some other diseases may have reduced the serum albumin levels or the levels may have still been recovering from diseases that have no obvious symptoms. With appropriate antimicrobial use (particularly carbapenems or aminoglycosides), the values had returned to their baseline levels by the day of discharge from the hospital (Fig. 1). We also investigated the incidence of *P. aeruginosa*, MRSA, and Candida as well as ESBL-producing *E. coli,* which are the most problematic bacteria in our hospital, but no clear relationship was observed between those bacteria and the low serum albumin levels. Therefore, it is likely that those low levels rather than the ESBL-producing *E. coli* infection, may have led the patients to become compromised hosts.

Lymphocytopenia is induced by protein-calorie malnutrition, immunosuppressive agents, cytotoxic chemotherapy, glucocorticoid therapy, viral infection, and protein-losing enteropathy.[29] However, both patients, whether infected or not with ESBL producers, showed no signs of lymphocytopenia (Table 1) and thus, the recent history of some diseases may have led patients to become compromised hosts, given that lymphocytes play a major immunological role in the prevention of infection.

CRP is an acute-phase protein whose level increases from 6 hours, reaching a peak at 48 hours after inflammation,[30] infection, surgeries, and burns.[31] In this study, because CRP level increased only in patients infected with ESBL-producing *E. coli* and decreased after recovery, that factor can directly reflect the infection state. It has been reported that inflammatory cytokines stimulate CRP production.[32] Whereas serum albumin levels and CRP levels exhibit a negative correlation in some diseases,[26,33,34] in this study we found no correlation between those two parameters (correlation coefficient: –0.019). In contrast, neutrophil levels and CRP levels show a strong correlation (correlation coefficient: 0.782).

Our results indicate that it is necessary to carefully monitor inpatients to determine whether or not they are infected with ESBL-producing *E. coli*, when they exhibit low serum albumin and lymphocyte levels. From the viewpoint of antimicrobial use, it is necessary early on to use carbapenems,[35] aminoglycosides, and fosfomycin.[3,36] However, it is also necessary to add the number of samples not only from our hospital but also other hospitals since the sample size may be small. Sometimes dietary intake, nutrient calories, and virus infection influence the albumin level and the number of lymphocytes, respectively. To confirm our hypothesis, we are planning to conduct basic research in the near future using animal models with hypoalbuminemia and hypolymphemia.

Furthermore, we must establish a system to share information concerning infections in the

community, since 48.0% of the patients who were transferred to our hospital were infected with ESBL-producing *E. coli* (unpublished data). Frequent communication and exchanges of information with community hospitals and clinics will be needed.[20,37]

Very recently, some carbapenemase-producing bacteria[38] and NDM-1 producers,[39-41] which are MDROs, have been reported. In order to prevent and minimize the spread of bacterial infections, predictions of infection and medical therapy as well as daily pathophysiological observations will be necessary.

Conflict of interest statement: None declared

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REFERENCES

1) Knothe H, Shah P, Krcmery V, Antal M, Mitsuhashi S. Transferable resistance to cefotaxime, cefoxitin, cefamandole and cefuroxime in clinical isolates of *Klebsiella pneumoniae* and *Serratia marcescens. Infection*, 1983; 11: 315–317.
2) Pitout JD, Laupland KB. Extended-spectrum beta-lactamase-producing Enterobacteriaceae: an emerging public-health concern. *Lancet Infect Dis*, 2008; 8: 159–166.
3) Rodríguez-Baño J, Alcalá JC, Cisneros JM, Grill F, Oliver A, Horcajada JP, Tórtola T, Mirelis B, Navarro G, Cuenca M, Esteve M, Peña C, Llanos AC, Cantón R, Pascual A. Community infections caused by extended-spectrum beta-lactamase-producing *Escherichia coli*. *Arch Intern Med*, 2008; 168: 1897–1902.
4) Ben-Ami R, Rodríguez-Baño J, Arslan H, Pitout JD, Quentin C, Calbo ES, Azap OK, Arpin C, Pascual A, Livermore DM, Garau J, Carmeli Y. A multinational survey of risk factors for infection with extended-spectrum beta-lactamase-producing enterobacteriaceae in nonhospitalized patients. *Clin Infect Dis*, 2009; 49: 682–690.
5) Hyle EP, Bilker WB, Gasink LB, Lautenbach E. Impact of different methods for describing the extent of prior antibiotic exposure on the association between antibiotic use and antibiotic-resistant infection. *Infect Control Hosp Epidemiol*, 2007; 28: 647–654. Review.
6) Urbánek K, Kolár M, Lovecková Y, Strojil J, Santavá L. Influence of third-generation cephalosporin utilization on the occurrence of ESBL-positive *Klebsiella pneumoniae* strains. *J Clin Pharm Ther*, 2007; 32: 403–408.
7) Nishio H, Sueyoshi N, Yamamoto K, Furukawa A, Takemura M, Yoshio K, Nakamura H, Inoue T. Laboratory-based surveillance of extended-spectrum β-Lactamase-producing *Escherichia coli*, *Klebsiella spp*, and *Proteus mirabilis*, by the Shiga Infection Control Network. *Jpn J Environ Infect*, 2009; 24, 170–176. (in Japanese)
8) Schwaber MJ, Navon-Venezia S, Kaye KS, Ben-Ami R, Schwartz D, Carmeli Y. Clinical and economic impact of bacteremia with extended-spectrum-beta-lactamase-producing Enterobacteriaceae. *Antimicrob Agents Chemother*, 2006; 50: 1257–1262.
9) Anderson DJ, Engemann JJ, Harrell LJ, Carmeli Y, Reller LB, Kaye KS. Predictors of mortality in patients with bloodstream infection due to ceftazidime-resistant *Klebsiella pneumoniae*. *Antimicrob Agents Chemother*, 2006; 50: 1715–1720.
10) Hyle EP, Lipworth AD, Zaoutis TE, Nachamkin I, Bilker WB, Lautenbach E. Impact of inadequate initial antimicrobial therapy on mortality in infections due to extended-spectrum beta-lactamase-producing enterobacteriaceae: variability by site of infection. *Arch Intern Med*, 2005; 165: 1375–1380.
11) Peña C, Gudiol C, Calatayud L, Tubau F, Domínguez MA, Pujol M, Ariza J, Gudiol F. Infections due to

Escherichia coli producing extended-spectrum beta-lactamase among hospitalised patients: factors influencing mortality. *J Hosp Infect*, 2008; 68: 116–122.

12) Tumbarello M, Sanguinetti M, Montuori E, Trecarichi EM, Posteraro B, Fiori B, Citton R, D'Inzeo T, Fadda G, Cauda R, Spanu T. Predictors of mortality in patients with bloodstream infections caused by extended-spectrum-beta-lactamase-producing Enterobacteriaceae: importance of inadequate initial antimicrobial treatment. *Antimicrob Agents Chemother*, 2007; 51: 1987–1994.
13) Johnson JR, Johnston B, Clabots C, Kuskowski MA, Castanheira M. *Escherichia coli* sequence type ST131 as the major cause of serious multidrug-resistant *E. coli* infections in the United States. *Clin Infect Dis*, 2010; 51: 286–294.
14) Barbe C, Fusellier A, Bureau Chalot F, Brasme L, Vernet Garnier V, de Champs C, Bajolet O. Predictive factors of acquisition of epidemic extended-spectrum beta-lactamase-producing *Escherichia coli*. *Pathol Biol (Paris)*, 2010; 58: 25–28.
15) Rodríguez-Baño J, Picón E, Gijón P, Hernández JR, Ruíz M, Peña C, Almela M, Almirante B, Grill F, Colomina J, Giménez M, Oliver A, Horcajada JP, Navarro G, Coloma A, Pascual A. Spanish Network for Research in Infectious Diseases (REIPI). Community-onset bacteremia due to extended-spectrum beta-lactamase-producing *Escherichia coli*: risk factors and prognosis. *Clin Infect Dis*, 2010; 50: 40–48.
16) Harris AD, McGregor JC, Johnson JA, Strauss SM, Moore AC, Standiford HC, Hebden JN, Morris JG, Jr. Risk factors for colonization with extended-spectrum beta-lactamase-producing bacteria and intensive care unit admission. *Emerg Infect Dis*, 2007; 13: 1144–1149.
17) Ofner-Agostini M, Simor A, Mulvey M, McGeer A, Hirji Z, McCracken M, Gravel D, Boyd D, Bryce E. Risk factors for and outcomes associated with clinical isolates of *Escherichia coli* and *Klebsiella species* resistant to extended-spectrum cephalosporins among patients admitted to Canadian hospitals. *Can J Infect Dis Med Microbiol*, 2009; 20: e43–48.
18) Clinical and Laboratory Standards Institute. 2006. Performance Standards for Antimicrobial Susceptibility Testing; 16th Informational Supplement. CLSI document M100-S16. Clinical and Laboratory Standards Institute, Wayne, PA.
19) Colodner R, Rock W, Chazan B, Keller N, Guy N, Sakran W, Raz R. Risk factors for the development of extended-spectrum beta-lactamase-producing bacteria in nonhospitalized patients. *Eur J Clin Microbiol Infect Dis*, 2004; 23: 163–167.
20) Ben-Ami R, Schwaber MJ, Navon-Venezia S, Schwartz D, Giladi M, Chmelnitsky I, Leavitt A, Carmeli Y. Influx of extended-spectrum beta-lactamase-producing enterobacteriaceae into the hospital. *Clin Infect Dis*, 2006; 42: 925–934.
21) Centers for Disease Control and Prevention. 2006. Management of multidrug-resistant organisms in healthcare settings.
http://www.cdc.gov/ncidod/dhqp/pdf/ar/mdroGuideline2006.pdf
22) Miró E, Mirelis B, Navarro F, Rivera A, Mesa RJ, Roig MC, Gómez L, Coll P. Surveillance of extended-spectrum beta-lactamases from clinical samples and faecal carriers in Barcelona, Spain. *J Antimicrob Chemother*, 2005; 56: 1152–1155.
23) Fuhrman MP. The albumin-nutrition connection: separating myth from fact. *Nutrition*, 2002; 18: 199–200.
24) Gibbs J, Cull W, Henderson W, Daley J, Hur K, Khuri SF. Preoperative serum albumin level as a predictor of operative mortality and morbidity: results from the National VA Surgical Risk Study. *Arch Surg*, 1999; 134: 36–42.
25) Don BR, Kaysen G, Serum albumin: relationship to inflammation and nutrition. *Semin Dial*, 2004; 17: 432–437. Review.
26) Kaysen GA. Biochemistry and biomarkers of inflamed patients: why look, what to assess. *Clin J Am Soc Nephrol*, 2009; 4: S56–63.
27) Margarson MP, Soni N. Serum albumin: touchstone or totem? *Anaesthesia*, 1998; 53: 789–803.
28) Oertel MF, Hauenschild A, Gruenschlaeger J, Mueller B, Scharbrodt W, Boeker DK. Parenteral and enteral nutrition in the management of neurosurgical patients in the intensive care unit. *J Clin Neurosci*, 2009; 16: 1161–1167.
29) Goldman L, Ausiello D. Cecil Medicine, 23rd Edition – Expert Consult Premium Edition. pp. 1258–1259, 2008, Saunders Elsevier Press, Philadelphia.
30) Pepys MB, Hirschfield GM. C-reactive protein: a critical update. *J Clin Invest*, 2003; 111: 1805–1812. Review.
31) Ho KM, Lipman J. An update on C-reactive protein for intensivists. *Anaesth Intensive Care,* 2009; 37: 234–241.
32) Heinrich PC, Castell JV, Andus T. Interleukin-6 and the acute phase response. *Biochem J*, 1990; 265:

621–636.

33) Kalender B, Mutlu B, Ersöz M, Kalkan A, Yilmaz A. The effects of acute phase proteins on serum albumin, transferrin and haemoglobin in haemodialysis patients. *Int J Clin Pract*, 2002; 56: 505–508.

34) Jones CH, Wolfenden RC, Wells LM. Is subjective global assessment a reliable measure of nutritional status in hemodialysis? *J Ren Nutr*, 2004; 14: 26–30.

35) Paterson DL, Ko WC, Von Gottberg A, Mohapatra S, Casellas JM, Goossens H, Mulazimoglu L, Trenholme G, Klugman KP, Bonomo RA, Rice LB, Wagener MM, McCormack JG, Yu VL. Antibiotic therapy for *Klebsiella pneumoniae* bacteremia: implications of production of extended-spectrum beta-lactamases. *Clin Infect Dis*, 2004; 39: 31–37.

36) Garau J. Other antimicrobials of interest in the era of extended-spectrum beta-lactamases: fosfomycin, nitrofurantoin and tigecycline. *Clin Microbiol Infect*, 2008; 14: 198–202.

37) Ostrowsky BE, Trick WE, Sohn AH, Quirk SB, Holt S, Carson LA, Hill BC, Arduino MJ, Kuehnert MJ, Jarvis WR. Control of vancomycin-resistant enterococcus in health care facilities in a region. *N Engl J Med*, 2001; 344: 1427–1433.

38) Nordmann P, Cuzon G, Naas T. The real threat of *Klebsiella pneumoniae* carbapenemase-producing bacteria. *Lancet Infect Dis*, 2009; 9: 228–236.

39) Kumarasamy KK, Toleman MA, Walsh TR, Bagaria J, Butt F, Balakrishnan R, Chaudhary U, Doumith M, Giske CG, Irfan S, Krishnan P, Kumar AV, Maharjan S, Mushtaq S, Noorie T, Paterson DL, Pearson A, Perry C, Pike R, Rao B, Ray U, Sarma JB, Sharma M, Sheridan E, Thirunarayan MA, Turton J, Upadhyay S, Warner M, Welfare W, Livermore DM, Woodford N. Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular, biological, and epidemiological study. *Lancet Infect Dis*, 2010; 10: 597–602.

40) Poirel L, Revathi G, Bernabeu S, Nordmann P. Detection of NDM-1-producing *Klebsiella pneumoniae* in Kenya. *Antimicrob Agents Chemother*, 2011; 55, 934–936.

41) Samuelsen O, Thilesen CM, Heggelund L, Vada AN, Kümmel A, Sundsfjord A. Identification of NDM-1-producing Enterobacteriaceae in Norway. *J Antimicrob Chemother*, 2011; 66: 670–672.