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Effect of Antibiotic Pretreatment on Resistance

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The emergence of bacterial resistance to antibiotics limits the efficacy of technical developments in the field of infectious diseases. This is particularly true for respiratory tract infections, which are by far the main reason for antibiotic use in developed countries. Antimicrobial resistance among respiratory pathogens involves both gram-positive (primarily *Streptococcus pneumoniae*) and gram-negative (*Haemophilus influenzae, Moraxella catarrhalis,* and the more rare enterobacteriaceae) microorganisms. A number of epidemiologic studies show a relationship between antibiotic use and antibiotic resistance, and how antibiotic pretreatment can reduce the range of effective drugs for optimal therapy of infections in general and of respiratory tract infections in particular. An appropriate use of antimicrobials is of crucial importance to limit the emergence and spread of bacterial resistance to antibiotics. This can be achieved by avoiding usage in nonspecific, probably viral, infections that are unlikely to be influenced by antibiotic therapy, and by using narrow-spectrum drugs to minimize selective pressure.

THE DISCOVERY OF potent antimicrobials has been a major medical progress during the second half of the 20th century. Unfortunately, the emergence of antimicrobial-resistant pathogens is limiting these advances. In particular, during the past 10 years, we witnessed a major shift in the etiology of both community- and hospital-acquired infections, from easy to treat pathogens toward more resistant pathogens, with fewer therapeutic options. The modified spectrum of pathogens and emerging bacterial resistance are changing the way in which infectious diseases are managed.[1]

Microbial resistance is the result of a number of causes and trends such as:

- Abuse and misuse of antimicrobials;
- Increased number of more susceptible hosts such as immune depressed patients, critically ill postsurgical and intensive care unit patients;
- Increased use of invasive procedures and devices;
- Occasional breakdown of infection control practices.[2]

It has been debated for many years whether bacterial resistance to antibiotics can influence clinical outcome. A relationship between *in vitro* susceptibility tests and antibiotic therapeutic failure can be supported by:

- Emergence, during therapy, of a new resistance marker not known previously;
- Selection of a resistant mutant or acquisition of a resistance gene during therapy;
- Failure to recognize or take into account new resistance mechanisms;
- Superinfection with resistant bacteria.[3]

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Emergence of antibiotic resistance among commonly encountered gram-positive cocci is currently a serious problem worldwide, the most important issues being penicillin-resistant *Streptococcus pneumoniae* (PRSP), vancomycin-resistant enterococci, methicillin-resistant *Staphylococcus aureus* (MRSA), and, most recently, glycopeptide-intermediate *S. aureus*. All these organisms are multidrug resistant.[4]

Another serious problem is antibiotic resistance of a number of gram-negative bacilli (*Klebsiella pneumoniae, Escherichia coli, Serratia marcescens*) owing to extended-spectrum β-lactamase production, and multiantibiotic resistance of *Pseudomonas aeruginosa*. All these pathogens are common members of the gastrointestinal flora and, therefore, pose potential problems in abdominal surgical infections, primarily in immune depressed patients. Because of their increasing dissemination, such resistant bacteria may have great clinical impact and make antimicrobial therapy of many infections extremely difficult or virtually impossible in some cases. The extensive, and often inappropriate, use of antibiotics worldwide has been known for a long time to be the major factor in the emergence and spread of antimicrobial resistance among microorganisms.[5]

Many investigators state that antibiotic-resistant bacterial strains arise primarily in hospitals, but this is only partly true. The widespread use of antibiotics by general practitioners, dermatologists, dentists, farmers, and so forth could be important for resistance development in the community as well. In addition, we must determine whether there are environmental selection pressures, other than antibiotic use, that could contribute to the spread of bacterial resistance and explain the high level of resistance in areas where antibiotics are not used. The key moment to stop resistance would be before it starts, by preventing the acquisition of resistance genes. From this point of view, long-term exposure to low antibiotic doses is the condition most likely to promote stable maintenance of resistance genes, whereas short-term exposure to antibiotic levels high enough to kill bacteria

or prevent their growth is much less likely to support resistance.[6]

Some investigators have discussed the role of antibiotic selective pressure behind the inappropriate use of antimicrobials. Every hospital should look at its own resistance situation and determine the best methods to control it.[2]

Today, it is necessary to predict the risk for antibiotic resistance and to understand how antibiotics are used and how their use can influence the evolution of resistance. Only if we understand the epidemiology of bacterial resistance to antimicrobials will we be able to implement preventive strategies to limit pre-existing resistance and to avoid the emergence of new resistant bacterial strains.[7] The correlation between antibiotic use and antibiotic resistance is not so easy to show, however, evidence is plentiful and mostly consistent. In the hospital, establishing an epidemiologic diagnosis is a prerequisite to any decision, including restriction of antibiotic use.[8]

To limit the prevalence of bacterial resistance to antimicrobials, 2 complementary strategies are of fundamental importance. The first one is to avoid antibiotics in very common situations in which they are unlikely to provide benefit, such as in a number of upper respiratory tract infections—common cold included—and in bronchitis. For example, it has been shown that 51% of patients diagnosed as having colds, 52% nonspecific upper respiratory tract infections, and 66% acute or nonspecific bronchitis, are treated by ambulatory care physicians with antibiotics.[9]

The second crucial point is to use narrow-spectrum antibiotics as much as possible to minimize selective pressure.[10] Much emerging evidence shows that these strategies can be effective.

In a child-care center in Omaha, NE, Boken et al[11] showed that nasopharyngeal carriage of PRSP decreased dramatically from 53% to 7% concomitantly with a decrease in antibiotic use by the attendees.

In Iceland, during the early 1990s, a nationwide campaign resulted in decreased antibiotic use by approximately 10%, with a reduction of as high as 30% for cotrimoxazole and macrolides. This campaign was followed by a significant decrease in the incidence of PRSP infections from 20% to 16.9%, and by a dramatic fall in the rate of carriage of PRSP from 49% to 15% among day-care center attendees.[12]

In Finland, a significant reduction in macrolide consumption from 2.40 defined daily doses per 1,000 per day in 1991 to 1.38 in 1992 ($P = .007$) was followed by a steady decrease in the frequency of erythromycin resistance among group A streptococci recovered from throat swabs and pus samples.[13]

North American guidelines recommending a reduction in the use of erythromycin and other macrolides for treatment of respiratory tract and skin and soft-tissue infections were instituted in the mid-1990s; shortly after, macrolide consumption decreased by 50%, and a similar reduction in the frequency of erythromycin-resistant isolates was reported.[10]

The incidence of nosocomial infections caused by gram-negative bacilli has been increasing lately. In the past decade, there was an increased use of third-generation cephalosporins (3GCs), both for first-line therapy and for prevention of infections in hospitalized patients. At the same time, resistance to 3GCs emerged among gram-negative bacilli capable of producing group I β-lactamases. These bacteria possess a gene that, when triggered by either exposure to 3GCs or spontaneous mutations, produces a cephalosporinase that can inactivate all of the currently available cephalosporins. *Citrobacter* spp., *S. marcescens, Enterobacter* spp., and *P. aeruginosa* are the most common gram-negative bacteria possessing this inducible enzyme.[14,15]

Moreover, there is the belief that the more and more frequent prescription of 3GCs partly results from their inappropriate use. In the mid-1990s, a multinational survey was performed on the type of antibiotic therapy prescribed by general practitioners of 7 European countries for therapy of community-acquired pneumonia, and the results of this study provided extremely varied information from country to country. Whereas in Sweden, the United Kingdom, and Holland, old, narrow-spectrum, oral antimicrobials (amoxicillin, erythromycin, oxytetracycline, doxycycline) were preferred, in Italy new parenteral 3GCs (ceftriaxone, ceftazidime, and cefotaxime) and carbapenems (imipenem) were mostly used.[16] It is obvious that such prescriptive behavior can have serious repercussions on both economic and ecologic grounds by producing a definite, significant increase in costs and a possible, dreadful spread of bacterial resistance.

Among other gram-negative bacteria that are important causes of respiratory tract infections, approximately one third of *Haemophilus influenzae* isolates and more than 90% of *Moraxella catarrhalis* strains in the United States produce β-lactamases.[17] The main consequence of this phenomenon is that such isolates are resistant to ampicillin and to other β-lactamase–sensitive penicillins, thus, requiring the use of more expensive and/or extended-spectrum antibiotics.

Medical literature supplies a number of reviews regarding the use of piperacillin-tazobactam for treatment of infections caused by β-lactamase–producing pathogens. Piperacillin alone has been successful in the therapy of many bacterial infections, however, it is susceptible to hydrolysis by many commonly encountered β-lactamases. The addition of tazobactam to piperacillin extends the spectrum of piperacillin, making it a valid alternative for treatment of nosocomial bacterial infections. Recently, the increased use of 3GCs has been associated with the emergence of resistant bacteria in the

nosocomial setting, including multiresistant enterococci and extended-spectrum β-lactamase–producing *Enterobacteriaceae*. An outbreak of ceftazidime-resistant *K. pneumoniae* (28% of all *K. pneumoniae* strains) at the Cleveland Veterans Affairs Hospital occurred where ceftazidime was administered most frequently. The outbreak was controlled by limiting the use of ceftazidime by approximately 50% and by using piperacillin-tazobactam as a broad-spectrum alternative: within a few months, isolation of ceftazidime-resistant *K. pneumoniae* decreased significantly (10.2%; $P < .05$).[18]

Regarding gram-positive cocci, the medical literature reports a reduction of vancomycin-resistant enterococci prevalence after the restriction of the use of cephalosporins and their substitution with broad-spectrum penicillins. Other control options include rational drug switching and cycling processes, though such practices need to be validated by controlled trials.[19] The spread of MRSA strains has become a major problem in many hospitals worldwide, and currently they account for 20% to 40% of all *S. aureus* nosocomial isolates wherever these strains are endemic, as in many areas of the United States and in Southern Europe.[20,21] Interestingly, well-designed studies have shown that the changeover from methicillin-susceptible to methicillin-resistant *S. aureus* can result from selective antibiotic pressure in hospitals after use of cephalosporins, primarily after cefazolin administration as presurgical prophylaxis.[22] Moreover, the circulation of resistant strains between health care facilities and the community is challenging control programs. As a consequence of the increasing rate of MRSA carriage in patients discharged from the hospital, MRSA infection is now commonly described in patients readmitted or even newly admitted from the community to the hospital.[23]

As far as respiratory tract pathogens are concerned, the epidemiology of community-acquired pneumonia has changed significantly in recent years, and emerging resistance among these microorganisms, particularly to β-lactams, is an increasing concern. Although emerging bacterial resistance has a variable impact among different countries, its growing importance is globally changing the classic therapeutic approach to both community- and hospital-acquired pneumonia. Today, a broad-spectrum empiric therapy is used most often for therapy of respiratory tract infections, and bacterial resistance needs be taken into account, though a direct relationship between antibiotic resistance and clinical outcomes in the treatment of pneumonia in adults has not been extensively shown. New antimicrobials must be considered, with emphasis on effective dosing and optimal dose interval.[1]

Penicillin resistance in *S. pneumoniae* is of 2 categories: intermediately susceptible isolates with minimal inhibitory concentrations comprised between 0.1 and 1 μg/L, and those with high-level resistance having minimum inhibitory concentrations greater than 1 μg/mL. The prevalence of these 2 types of resistance varies considerably, not only by geographic region but also by age and anatomic source. Overall in the United States, the percentage of penicillin-nonsusceptible *S. pneumoniae* (PNSP) is 34.9% (22.1% of organisms are intermediately susceptible, and 12.8% highly resistant), varying from 28% in North- and Mid-Atlantic states to 44% in South-Atlantic states.[17] Pneumococcal isolates from the upper respiratory tract seem to be more penicillin resistant than those from other sites (blood, sputum, eye): the incidence of PNSP was 53.7% for nasopharyngeal, 58.2% for middle ear, and 60.9% for sinus isolates. Patients younger than 2 years with otitis media had the highest prevalence of PNSP. High-level penicillin resistance is also commonly associated with multidrug resistance. In a recent review of 1,476 pneumococcal strains isolated in regional laboratories, macrolide resistance was found in 30% of all strains, but in 67% of isolates that were highly resistant to penicillin.[24]

At present, penicillin continues to be the treatment of choice for pneumococcal pneumonia—even if caused by intermediately susceptible strains—for a number of reasons such as high penetration into the respiratory tract, low cost, and low selective pressure. In the ambulatory setting, empiric use of other agents such as macrolides, fluoroquinolones, or doxycycline can be a reasonable choice.[25]

A number of studies have been published in medical literature about the relationship between prior antibiotic use and emergence of resistant bacterial isolates. Spanish researchers have recently published worrisome data on the frequency of PNSP as an agent of acute otitis media in their country. During the study period, they noted a significant difference ($P = .01$) between the isolation rate of PNSP from children with acute otitis media not recently treated (50%) and recently treated (90.5%) with antibiotics, and the same trend, though not significant, was noted for strains that were resistant to erythromycin (35.7% and 62%; $P = .12$), clindamycin (35.7% and 47.6%; $P = .48$), and cotrimoxazole (62.4% and 81%; $P = .23$). The rates of β-lactamase–producing *H. influenzae* isolates were different between treated (43%) and not treated (23%) patients as well, though the difference was not statistically significant ($P = .18$). The investigators concluded that isolation of PNSP from patients with recurrent acute otitis media suggests an important role of the selection of resistant mutants resulting from prior antibiotic use.[26]

Other reports have also shown high rates of PNSP in respiratory tract infections that do not respond to initial antibiotic therapy, and PNSPs were found several times more frequently in recurrent acute otitis media than in acute untreated infections. For example, Block et al[27] isolated PNSPs much more commonly from patients recently treated (within 3 days) with antibiotics (30%) than

from patients treated more than 3 days earlier (2%; $P < .0001$). If one considers the annual high rate of antimicrobial prescriptions for nonspecific upper respiratory tract infections, bronchitis, and even the common cold,[9] it is obvious that efforts to improve awareness in patients and/or parents, physicians, and public health officials and to limit unnecessary antibiotic use for treatment of respiratory tract infections have to be made.[28]

In the mid 1990s, Jacobson et al[14] performed a prospective, case-controlled, observational study to examine the prevalence and the risk factors for development of resistance to extended-spectrum cephalosporins (ESCs) in group I β-lactamase–producing gram-negative bacilli. A total of 386 isolates were recovered from 340 patients during the study period, and 70 (18.1%) of them were resistant to 3GCs, with strong differences among both genera and species. For instance, a good 31.1% of *E. cloacae* and only 18.9% of *E. aerogenes* isolates were resistant. *P. aeruginosa* was the second most frequently isolated organism after *Enterobacter* spp. (40.2% of all isolates), but with a much lower resistance rate (7.7%). *Citrobacter* spp. represented only 8% of all isolates, and all but one of the resistant *Citrobacter* isolates were of the species *C. freundii,* which had a resistance rate of 40.9%. *S. marcescens* represented 9.1% of all isolates and had a 5.7% resistance rate.[14] In this study, the investigators concluded that:

- The mean number of antibiotics used by patients before isolation of a resistant isolate was significantly greater than among patients with a susceptible strain. Twenty-eight percent of patients whose isolates were susceptible had not received an antibiotic during the prior 30 days, but only 7.6% of the patients whose isolates were resistant had not received an antibiotic beforehand.
- Resistant isolates were recovered less frequently from patients receiving a 3GC plus an aminoglycoside than from those receiving a 3GC alone, thus, suggesting that bacterial resistance could be markedly lower if aminoglycosides are associated with 3GCs.
- The number of days of antibiotic therapy before the emergence of resistance was compared with the probability of development of resistance. A linear relationship between the number of days of therapy with ceftizoxime or cefotaxime and the probability of development of resistance was noted. However, even 1 day of therapy with ceftazidime put patients at risk for having an isolate resistant to 3GCs.

They also found:

- A strong correlation between the use of broad-spectrum antibiotics and the subsequent isolation of group I β-lactamase–producing organisms that are resistant to multiple ESCs. These multiresistant strains are more and more frequently causing infections in hospitalized patients and increasing the mortality rate. In particular, prior exposure to ceftizoxime, cefotaxime, ceftazidime, and, probably, piperacillin was associated with the isolation of organisms resistant to 3GCs. This finding argues for selection, rather than induction, of more resistant endemic hospital flora as the mechanism responsible for emergence of resistance after piperacillin use.
- An association between the isolation of resistant *Enterobacter* strains and prior therapy with 3GCs. Resistant bacteria have been isolated from up to 44% of patients infected with *Enterobacter* spp. and treated with 3GCs.[14] Many reports have described clinical failure related to this phenomenon. The presence of multiresistant *Enterobacter* spp. has also been associated with a longer hospital stay, greater use of antibiotics, and a higher mortality rate.[29] Other underlying conditions (ie, neutropenia and cystic fibrosis) have been found to be associated with the emergence of pathogens resistant to 3GCs.[30]

We can find a number of reports regarding the emergence of microorganisms resistant to multiple β-lactams during therapy with 3GCs because of the production of β-lactamases. A recent prospective study on the emergence of resistance during therapy for *Enterobacter* bacteremia showed that previous administration of ESCs was more likely to be associated with recovery of multiresistant *Enterobacter* isolates in an initial positive blood culture (69%) than was administration of antibiotics that did not include an ESC (20%).[29] A 6-month prospective investigation studied 134 patients from whom *Enterobacter* isolates were recovered: the rate of resistance was 25.4%, and the prior use of a 3GC was significantly associated with resistance ($P < .0001$).[31]

One method by which emergence of resistance can be prevented, or at least delayed, is the use of combination therapy. Because the emergence of resistant mutants is the result of selective pressure by antimicrobial therapy, the change of mutants resistant to 2 antimicrobials in the parent population being present is a product of mutation frequencies, provided that resistance mechanisms are independent. From both *in vitro* and *in vivo* studies it seems possible that emergence of resistance is less common when combination therapy is used, primarily for microorganisms able to develop resistance relatively quickly, such as *P. aeruginosa,* and for resistance mechanisms that occur at a relatively high frequency.[32]

A number of reports document conflicting results on the combination therapy with a 3GC plus an aminoglycoside in preventing the emergence of resistance to 3GCs, though most series included small numbers of patients. Chow et al[29] showed that the emergence of resistance to 3GCs was not less common among patients receiving a 3GC plus an aminoglycoside than among those receiving a 3GC alone. On the other hand, the aforementioned study by Jacobson et al,[14] using logistic regression analysis, found a significant reduction in the development of resistance when an

aminoglycoside was added. Although the use of an aminoglycoside plus a 3GC does not always prevent the emergence of resistance, the organisms usually remain susceptible to 1 of the agents, thus, possibly improving the chances of patients' survival.[14]

An important part of the strategies aimed at circumventing the development of bacterial resistance is to assess the ecologic impact of preventive antibiotic therapy in hospital practice.[33] More information is needed to better define mechanisms of resistance and to clarify more effective ways of using existing antimicrobials. Appropriate use of antibiotics in the hospital (primarily in the intensive care unit, in severely ill patients, and for complicated infections) must to be optimized to reduce the emergence of further resistance. It is anticipated that preventive and therapeutic options will provide the basis for a better approach to this problem. The consequences associated with antimicrobial resistance should serve as a strong incentive to establish appropriate and judicious use of antibiotics.[2]

REFERENCES

1. Lister PD: Emerging resistance problems among respiratory tract pathogens. Am J Manag Care 6:409-418, 2000 (suppl)
2. File TM: Emerging antimicrobial resistance on the eve of the new millennium. Hospital practice, a special report: Challenges in the management of antimicrobial-resistance pathogens. Minneapolis, McGraw-Hill, 1998
3. Acar JF, Goldstein FW: Consequences of increasing resistance to antimicrobial agents. Clin Infect Dis 27:125-130, 1998 (suppl)
4. Hand WL: Current challenges in antibiotic resistance. Adolesc Med 11:427-438, 2000
5. McGowan JE Jr: Antimicrobial resistance in hospital organisms and its relation to antibiotic use. Rev Infect Dis 5:1033-1048, 1983
6. Salyers AA, Amabile-Cuevas CF: Why are antibiotic resistance genes so resistant to elimination? Antimicrob Agents Chemother 11:2321-2335, 1997
7. Guillemot D: Antibiotic use in humans and bacterial resistance. Curr Opin Microbiol 2:494-498, 1999
8. Monnet DL: Antibiotic use and bacterial resistance. Ann Fr Anesth Reanim 19:409-417, 2000
9. Gonzales R, Steiner JF, Sande MA: Antibiotic prescribing for adults with colds, upper respiratory tract infections, and bronchitis by ambulatory care physicians. JAMA 278:901-904, 1997
10. Bennett J, Geme JW: Bacterial resistance and antibiotic use in the emergence department. Pediatr Clin North Am 46: 1125-1143, 1999
11. Boken DJ, Chartrand SA, Goering RV, et al: Colonization with penicillin-resistant *Streptococcus pneumoniae* in a child-care center. Pediatr Infect Dis J 14:879-884, 1995
12. Kristinsson KG, Hjalmarsdottir MA, Gudnason TH: Continued decline in the incidence of penicillin nonsusceptible pneumococci in Iceland [abstract C-22]. Abstracts of the 38th Interscience Conference on Antimicrobial Agents and Chemotherapy, San Diego, CA, 1998, pp 74-75
13. Seppälä H, Klaukka T, Vuopio-Varkila J, et al: The effect of changes in the consumption of macrolide antibiotics on erythromycin resistance in group A streptococci in Finland. N Engl J Med 337:441-446, 1997
14. Jacobson KL, Cohen SH, Inciardi JF, et al: The relationship between antecedent antibiotic use and resistance to extended-spectrum cephalosporins in group 1 β-lactamase-producing organisms. Clin Infect Dis 21:1107-1113, 1995
15. Pechère JC: Emergence of resistance in gram-negative bacilli during β-lactam therapy: A challenge for the future. Eur J Cancer 25:17-23, 1989 (suppl)
16. Örtqvistå: Antibiotic treatment of community-acquired pneumonia in clinical practice: A European perspective. J Antimicrob Chemother 35:205-212, 1995
17. Thornsberry C, Jones ME, Hickey ML, et al: Resistance surveillance of *Streptococcus pneumoniae, Haemophilus influenzae* and *Moraxella catarrhalis* isolated in the United States, 1997-1998. J Antimicrob Chemother 44:749-759, 1999
18. Rice LB, Eckstein EC, DeVente J, et al: Ceftazidime-resistant *Klebsiella pneumoniae* isolates recovered at the Cleveland Department of Veterans Administration Affairs Medical Center. Clin Infect Dis 23:118-124, 1996
19. Shlaes DM, Gerding DN, John JF Jr, et al: Society for Healthcare Epidemiology of America and Infectious Diseases Society of America Joint Committee on the prevention of antimicrobial resistance: Guidelines for the prevention of antimicrobial resistance in hospitals. Clin Infect Dis 25:584-599, 1997
20. Fridkin SK, Steward CD, Edwards JR, et al: Surveillance of antimicrobial use and antimicrobial resistance in United States hospitals: Project ICARE phase 2. Clin Infect Dis 29:245-252, 1999
21. Voss A, Milatovic D, Wallrauch-Schwarz C, et al: Methicillin- resistant *Staphylococcus aureus* in Europe. Eur J Clin Microbiol Infect Dis 13:50-55, 1994
22. Schentag JJ, Hyatt JM, Carr JR, et al: Genesis of methicillin-resistant *Staphylococcus aureus* (MRSA), how treatment of MRSA infections has selected for vancomycin-resistant *Enterococcus faecium*, and the importance of antibiotic management and infection control. Clin Infect Dis 26:1204-1214, 1998
23. Girou E, Pujade G, Legrand P, et al: Selective screening of carriers for control of methicillin-resistant *Staphylococcus aureus* (MRSA) in high-risk hospital areas with a high level of endemic MRSA. Clin Infect Dis 27:543-550, 1998
24. Jacobs MR, Bajaksouzian S, Lin G, et al: Variation in antimicrobial resistance of community isolates of *Streptococcus pneumoniae* by age, region, and disease [abstract C-16]. Abstracts of the 38th Interscience Conference on Antimicrobial Agents and Chemotherapy, San Diego, CA, 1998, p 73
25. Harwell JI, Brown RB: The drug resistance pneumococcus, clinical relevance, therapy, and prevention. Chest 117:530-541, 2000
26. Del Castillo F, Baquero-Artigao F, Garcia-Perea A: Influence of recent antibiotic therapy on antimicrobial resistance of *Streptococcus pneumoniae* in children with acute otitis media in Spain. Pediatr Infect Dis J 17:94-97, 1998
27. Block SL, Harrison CJ, Hedrick JA, et al: Penicillin-resistant *Streptococcus pneumoniae* in acute otitis media: Risk

factors, susceptibility patterns and antimicrobial management. Pediatr Infect Dis J 14:751-759, 1995

28. Schwartz B, Bell DM, Hughes JM: Preventing the emergence of antimicrobial resistance: A call for action by clinicians, public health officials, and patients. JAMA 278:944-945, 1997

29. Chow JW, Fine MJ, Shales DM, et al: *Enterobacter* bacteremia: Clinical features and emergence of antibiotic resistance during therapy. Ann Intern Med 115:585-590, 1991

30. Dworzak DL, Pugsley MP, Sanders CC, at al: Emergence of resistance in gram-negative bacteria during therapy with expanded-spectrum cephalosporins. Eur J Clin Microbiol 6: 456-459, 1987

31. Cohen SH, King JH, Morita MM, et al: The relationship between *Enterobacter* isolates resistant to extended spectrum cephalosporins and antecedent antibiotic use [abstract 654]. Program and Abstracts of the 29th Interscience Conference on Antimicrobial Agents and Chemotherapy, Houston, TX, 1989

32. Mouton JW: Combination therapy as a tool to prevent emergence of bacterial resistance. Infection 27:24-28, 1999 (suppl)

33. Korinek AM: Ecological consequences of preventive antibiotic prescriptions. Ann Fr Anesth Reanim 19:418-423, 2000