

# EFFECT OF CEFOXITIN AND CLINDAMYCIN ON SELECTION OF DEREPRESSED MUTANTS IN ENTEROBACTER CLOACAE

D. Saverino<sup>1</sup>, F. Milintenda-Floriani<sup>1</sup>, A.A. Medeiros<sup>2</sup>

<sup>1</sup>Department of Experimental Medicine, Section of Human Anatomy, University of Genova, Italy;

<sup>2</sup>The Miriam Hospital, Brown University, Providence R.I. (USA)

## SUMMARY

The characteristics of an antibiotic that favor its ability to select for resistant bacteria are not completely understood. Otherwise, by the common use of broad-spectrum cephalosporins, resistant strains of several gram-negative species, especially *Enterobacter cloacae*, have been more frequently isolated. During our studies on  $\beta$ -lactam resistance in *E. cloacae*, we observed that the addition of an inhibitor (clindamycin) to a potent inducer (cefoxitin) leads to an enhanced selection of resistant mutants. This could explain the emergence of  $\beta$ -lactam resistant strains during antibiotic therapy.

**KEY WORDS:**  $\beta$ -lactamase, inducibility, *Enterobacter cloacae*, cefoxitin, clindamycin

Received September 15, 2003

Accepted March 20, 2004

## INTRODUCTION

The development of resistance of gram-negative bacteria to  $\beta$ -lactam antibiotics during therapy has been well described (Beckwith *et al.*, 1980; Black *et al.*, 1985; Blahova *et al.*, 1996; Livermore *et al.*, 1996; Palmer, 1995; Sanders *et al.*, 1983). Some species such as *Enterobacter cloacae* and *Pseudomonas aeruginosa*, in which this occurred produce inducible, chromosomally encoded Class I  $\beta$ -lactamases (Pfaller *et al.*, 1997; Seeberg *et al.*, 1983; Vu *et al.*, 1985). The resistant variants usually are stably derepressed mutants that hyper-produce the  $\beta$ -lactamase.

Some authors suggest that antibiotics with poor inducer activity are the preferred agents for infections with organisms possessing inducible  $\beta$ -lactamases (Cavalieri *et al.*, 1991; Sanders, 1984; Weber *et al.*, 1990; Weinbren *et al.*, 1986). However, others have suggested that since induction causes only a transient increase in  $\beta$ -lactamase production, the antibiotic is not responsible for clinically relevant resistance

(Culmann *et al.*, 1985; Watanabe *et al.*, 1992). Rather, the principal determinant of resistance in clinical isolates may be the ability of an antibiotic to permit growth of pre-existing stably derepressed mutants (Cavalieri *et al.*, 1991; Livermore, 1986; Weber *et al.*, 1990) while inhibiting the more susceptible parent organisms. In theory, a  $\beta$ -lactam antibiotic that is a good inducer will foster its own destruction, thus limiting its ability to select resistant mutants.

In this paper we analyzed the efficacy of cefoxitin, a potent inducer (Sanders *et al.*, 1979), to select for stably derepressed mutants, both in the presence and absence of clindamycin, a known inhibitor of the induction of the chromosomal  $\beta$ -lactamase of *E. cloacae* (Sanders *et al.*, 1983).

## MATERIALS AND METHODS

### *Bacteria*

*Enterobacter cloacae* 3003 is a clinical isolate. *E. cloacae* 3003R, its cefotaxime-resistant mutant,