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Possible Link between Bacterial Resistance and Use of Antibiotics and Biocides

Low-level plasmid-mediated resistance to cationic biocides such as chlorhexidine (CHX), quaternary ammonium compounds (QACs), amidines, and acridines has been observed in antibiotic-resistant strains of *Staphylococcus aureus* and *Staphylococcus epidermidis* (1, 4, 5), and it has been postulated that strains in which *qac* genes are present might have enhanced survival in the clinical environment. Extensive use of cationic biocides could lead to the selection of staphylococcal strains showing resistance to both antibiotics and biocides (9, 11), but the clinical relevance of this possibility remains contentious (8).

Although plasmid-mediated resistance to biocides has also been found in gram-negative bacteria, it has been proposed (8) that intrinsic resistance in these organisms is of greater significance. Resistance to both antibiotics and biocides in gram-negative organisms is more likely where less specific mechanisms are involved, e.g., the outer membrane may act as a nonspecific exclusion blanket thereby preventing the uptake of chemically unrelated molecules (10, 11). There have, however, been some instances where biocides have been claimed to select for resistant gram-negative bacteria. Stickler et al. (12) observed resistance to CHX, QACs and at least five antibiotics for gram-negative bacteria isolated from urinary tract infections and proposed that the widespread use of CHX was responsible for selecting antibiotic-resistant strains. There was no evidence of plasmid-linked resistance association (although the possibility was raised but not proven that CHX-resistant strains were acting as more efficient recipients of plasmids conferring antibiotic resistance). Dance et al. (2) isolated a strain of *Proteus mirabilis*, responsible for a hospital outbreak, that was resistant to CHX and antibiotics. However, there was no evidence of a genetic linkage between these resistances, which were considered to be intrinsic rather than plasmid mediated. Selection of antibiotic-resistant bacteria by chlorination of drinking water and treated sewage has also been described, and the basis of linked biocide-antibiotic resistance in organisms isolated from aquatic sources is of potential public health importance (9).

TABLE 1. Responses of CHX-susceptible and CHX-resistant strains of *P. stutzeri* to other antibacterial agents

Antibacterial agent[a]	MIC (μg/ml) for *P. stutzeri*[b]					
	568	568R	10783	10783R	JM 302	JM 302R
CHX	2.5	10	2.5	50	2.5	100
CPC	25	100	25	250	50	100
TC	1	250	1	1	1	25
PB	<1	>500	<1	<1	<1	200
GM	2.5	5	<1	<1	<1	100
NA	5	100	25	25	10	10
EM	25	>200	25	>200	50	5
AM	10	>500	100	100	50	300

[a] CPC, cetylpyridinium chloride; TC, triclosan; PB, polymyxin B sulfate; GM, gentamicin; NA, nalidixic acid; EM, erythromycin; AM, ampicillin.
[b] R denotes CHX-resistant strain.

We have developed stable CHX resistance in some strains of *Pseudomonas stutzeri* by exposure to increasing concentrations of the bisbiguanide. MICs of CHX for parent strains were 2.5 to 5 μg/ml and MICs for resistant strains were 10 to 100 μg/ml. The CHX-resistant strains showed a variable increase in resistance to QACs (benzalkonium chloride and cetylpyridinium chloride) and to triclosan (increases in MICs of the phenolic of 25- to 250-fold). Additionally, these CHX-resistant strains also demonstrated a variable increase in resistance to polymyxin B, gentamicin, nalidixic acid, erythromycin, and ampicillin (Table 1). Resistant cells took up less CHX from solution than susceptible cells and cell envelope changes were observed microscopically, implicating the outer membrane as being involved in this reduced susceptibility.

Moken et al. (6) have described the selection of low-level chromosomal antibiotic resistance in *Escherichia coli* following exposure to a sublethal concentration of pine oil and have demonstrated that an export resistance mechanism (3) was involved. They also found that deletion of *acrAB*, but not of *mar*, resulted in a >10-fold increase in susceptibility of strains to a QAC and to chloroxylenol and thereby surmised that *acrAB* was also involved in efflux of these two biocides.

Concern about a possible linkage between antibiotic and biocide resistance has again been expressed recently (7) and clearly needs to be considered further in a clinical context. It is likely that more than one type of mechanism, viz., outer membrane changes and efflux, is involved.

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