1-AMINOETHYLPHOSPHONIC ACID, AN INHIBITOR OF BACTERIAL CELL WALL SYNTHESIS

Sir:

There are numerous steps in bacterial cell wall synthesis[11] but the compounds inhibiting this synthesis are limited. To this group of compounds, *i.e.* penicillins[2], cephalosporins[2], ristocetin[6], vancomycin[7], bacillin[1], moenomycin[5], prasinomycin[9], cycloserine[10], O-carbamyl-D-serine[10], glycine[8], and D-amino acids[3] can be added 1-aminoethylphosphonic acid (AEPA).

When titrated against *Proteus vulgaris*, using previously described methods[1], AEPA caused the same visible structural changes in the cells as did increasing concentrations of a classical cell wall antibiotic such as penicillin. Cells grown in presence of approximately 100 μg/ml of AEPA are much elongated and enlarged in girth and at somewhat higher concentrations the elongated, enlarged cells become herniated. The hernias become larger with increasing concentration of compound until at 750 μg/ml most of the cells are large spheroplasts. The lowest concentrations of antibiotics which induce cells of *Proteus vulgaris* to become spheroplasts are summarized in Table 1. As the DL form of AEPA was used in the experiments, the compound may be twice as effective if only one isomer is active. Two related compounds, 2-aminoethylphosphonic acid and aminomethylphosphonic acid, were inacitive.

Table 1. Comparative activity of compounds against *Proteus vulgaris*

Compound	Spheroplast inducing concentration* μg/ml
Penicillin G	5
Cephalosporin C	5
Cycloserine	150
Ristocetin	1,000
Vancomycin	2,000
1-Aminoethylphosphonic acid	750

* Concentration which causes greater than 99 percent of cells to form spheroplasts.

This strain of *Proteus vulgaris* is very sensitive to AEPA. Approximately 1 mg/ml of the compound is required to induce spheroplast formation in a strain of *Escherichia coli* and a strain of *Vibrio percolans*.

AEPA potentiates the action of penicillin and cycloserine. When *Proteus vulgaris* is grown in medium containing AEPA plus penicillin or AEPA plus cycloserine, using concentrations, which alone cause no observable effect on cell morphology, marked changes leading to spheroplast formation can be seen.

D-Alanine reverses the effect of cycloserine on spheroplast formation[10] and has a similar effect in combination with AEPA. When used at 2 mg/ml, D-alanine completely blocked the action of 1 mg/ml of AEPA. Though the mode of action of AEPA has not been determined, its structural similarity to alanine plus its reversal, and that of cycloserine, by D-alanine as well as its potentiation of cycloserine indicates that it may act at the steps inhibited by cycloserine[10]. Thus its site of action probably is different from that of phosphonomycin[4], another phosphonic acid compound subsequently discovered. In addition, combinations of AEPA and phosphonomycin do not induce formation of good spheroplasts. The cells become enlarged and distorted but the morphology is quite different from that obtained by use of AEPA or phosphonomycin alone.

Unlike phosphonomycin, AEPA has shown no *in vivo* activity in the infections studied to date (personal communication from Dr. A. K. MILLER).

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