In Vitro Interactions between β-Lactam Antibiotics and Tobramycin

To the Editor:

Aminoglycosides such as gentamicin and tobramycin are often used in combination with a β-lactam antibiotic, either to provide a wider spectrum of activity or for synergy against *Pseudomonas aeruginosa* and various Enterobacteriaceae (1). Monitoring aminoglycoside concentrations in serum has become an accepted therapeutic procedure in patients being treated with these agents because of the drugs' narrow therapeutic range.

Aminoglycosides are known to react chemically with certain β-lactam antibiotics and consequently to lose their antibacterial activity (2, 3). Much of the research into this reaction has been directed towards defining its significance in vivo (4, 5). Very few data exist that might aid the clinical laboratory in handling the samples collected for aminoglycoside assay from patients also receiving a β-lactam drug.

To assess the potential in vivo significance of this interaction, we prepared samples of human sera containing tobramycin (10 mg/L) and the following β-lactam antibiotics: carbenicillin (200 mg/L), ticarcillin (200 mg/L), penicillin G (75 mg/L), ampicillin (100 mg/L), mezlocillin (200 mg/L), cefalothin (75 mg/L), cefazolin (150 mg/L), cefoxitin (75 mg/L), or moxalactam (100 mg/L). These are the maximum concentrations likely to be encountered in clinical samples. Samples were assayed immediately for tobramycin concentration, left at room temperature for 48 h and then re-assayed by radioimmunoassay (RIANEN; New England Nuclear, Boston, MA 02118). The assay measures only active tobramycin (6) and has a coefficient of variation of 7.1%.

We considered losses of tobramycin activity of greater than 10% over 48 h to be clinically significant; of the drugs tested, only ticarcillin and carbenicillin caused this much loss of activity. We then carried out further studies with ticarcillin and carbenicillin, mixing tobramycin (9 mg/L) with 25, 50, 100, or 200 mg of ticarcillin or carbenicillin per liter in human sera. Immediately after mixing, each sample was assayed for tobramycin. Each mixture was then divided into three portions, one of which was left at room temperature, one refrigerated, and one frozen. In addition, to a portion of the 200 mg/L mixture containing carbenicillin we added penicillinase (Becton Dickinson & Co., Rutherford, NJ 07070) to a final concentration of 50 mega-kinetic (Kinsey) units/L and then refrigerated the sample. Each sample was assayed for tobramycin activity at 24, 48, 72, and 96 h after mixing.

There was significant inactivation of tobramycin with either ticarcillin or carbenicillin at concentrations of 50 mg/L or greater, the degree of inactivation increasing with increasing concentration. At room temperature, the tobramycin half-life was 67 h with carbenicillin (200 mg/L) and 101 h with ticarcillin (200 mg/L). Figure 1 illustrates the time course of tobramycin activity for mixtures containing 200 mg of carbenicillin per liter. Loss of activity exceeding 10% is apparent as early as 12 h when samples are kept at room temperature. Refrigeration slows the reaction but does not stop it, a 10% loss of activity occurring after about 24 h. Adding the penicillinase to the sample and refrigeration appears to slow the reaction effectively. Frozen samples had no significant loss of activity.

These results have important implications for the laboratory determining tobramycin concentrations in serum. Samples should not be left at room temperature for more than a few hours and should be sent to the laboratory as soon as possible. If refrigerated, samples should be assayed within 24 h of collection. In situations where this is not possible, we recommend either freezing the samples or adding at least 50 mega-units of penicillinase per liter. We would suggest these precautions be taken with all samples for tobramycin assay unless the laboratory has a reliable procedure for determining whether the patient is receiving carbenicillin or ticarcillin concomitantly.

References

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Aluminum Foil Instead of Glass Plates for Thin-layer Chromatography in Radioenzymic Assay

To the Editor:

The assay method we use in our laboratory for norepinephrine, epinephrine, and dopamine in plasma is essentially that described by Peuler and Johnson (1), which is commercially available in kit form ("Cat-A-Kit"; Upjohn Diagnostics).

We have made some modifications concerning the thin-layer chromatographic (TLC) step. Instead of glass plates, we use TLC aluminum sheets