Antibacterial Properties of 3,3',5,5'-Tetrachlorotetranitrodiphenylamine


Department of Chemistry, Loyola University of Chicago, Chicago, Illinois 60626, and U. S. Naval Dental Research Institute and Medical Research Unit No. 4, Great Lakes, Illinois 60088

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An analogue of tetramethylidicyramine, 3,3',5,5'-tetrachlorotetranitrodiphenylamine, was synthesized and tested in vitro for its antibacterial activity.

Tetramethylidicyramine, originally synthesized as an analytical reagent (6), has been reported to have antibacterial (3, 5) and antiviral properties in vitro (M. J. Rosenbaum et al., Antimicrob. Agents Chemother., in press). Analogs of tetramethylidicyramine were prepared to determine if they possessed better antibacterial properties than the parent agent. One such compound, 3,3',5,5'-tetrachlorotetranitrodiphenylamine, had excellent antibacterial activity and is reported here. The compound was synthesized from 3,3',5,5'-chlorodiphenylamine which was originally prepared by the Chapman rearrangement (1). The material was then nitrated under mild conditions to give a compound whose chemical analysis and spectral information indicated that it was most likely a 2,2',6,6'-tetraniitated product, 3,3',5,5'-tetrachlorotetranitrodiphenylamine. This compound was tested in vitro for its antibacterial activity.

The antibacterial action of the compound was tested on the following organisms: Staphylococcus aureus $\#198$, 201, and 204 (NDRI strains), Streptococcus species HS-6 ATCC $\#19642$, Streptococcus species FA-1 ATCC $\#19645$, Streptococcus sanguis species (SBE), Bacillus subtilis ATCC $\#9372$, Lactobacillus plantarum ATCC $\#8014$, Pseudomonas aeruginosa, Proteus vulgaris, Aerobacter aerogenes, and Herellea vaginicola. The yeastlike fungus Candida albicans was also tested.

The organisms were grown on Todd-Hewitt broth solidified with 1.5% agar (Difco), and supplemented with 1% dextrose. Filter paper discs (Schleicher & Schuell Co., Keene, N.H.; 1 $\#740-4$, diameter 12.7 mm) were impregnated with the compound at a concentration of 10 $\mu$g/ml and placed in the inoculated petri dishes for 24 hr at 37 C when the zone of inhibition was recorded. The B. subtilis and staphylococci strains gave the largest zones of inhibition (average 17.5 mm), while the streptococci species and the L. plantarum produced the smallest zones of inhibition (average 14.5 mm). The C. albicans and the gram-negative organisms were not inhibited.

The bacteriostatic concentration of the compound for S. aureus $\#198$, B. subtilis ATCC $\#9372$, and Streptococcus species FA-1 ATCC $\#19645$ was tested using the tube dilution technique. The tubes without growth were subcultured in Todd-Hewitt broth to determine the bactericidal concentration.

The minimal bacteriostatic concentration of the compound for S. aureus and B. subtilis was 0.156 $\mu$g/ml. The bactericidal concentration was 0.312 $\mu$g/ml. The compound was bacteriostatic for Streptococcus species FA-1 at 5.0 $\mu$g/ml and bactericidal above this level.

Eighteen sulfadiazine-resistant and 12 sulfadiazine-sensitive strains of Neisseria meningitidis of groups B, C, and Y(4) were also tested. The assay was carried out on Mueller-Hinton agar by the method of Frank, Wilcox, and Finland (2). The minimal inhibitory concentration of this compound ranged from 0.078 to 0.156 $\mu$g of medium per ml for both the sulfadiazine-resistant and sensitive groups.

Nitration of tetrachlorodiphenylamine under forcing conditions apparently produces a dihydroxy product possessing high antibacterial activity against gram-positive bacteria and the N. meningitidis group.

LITERATURE CITED