Determining the Mechanism of Action of Three Novel Chemical Scaffolds with Broad-Spectrum Antibacterial Activity, including Multidrug-resistant Gram-Negative Pathogens

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ABSTRACT

Objectives: Three completely novel classes of protein synthesis inhibitors have been designed and prepared as broad-spectrum agents, focused on modulating multidrug-resistant (MDR) Gram-negative pathogens. Each class evolved from a unique scaffold design, the target was the ribosome small subunit 50S. A key discovery was that the Gram-negative pathogens were attributable to protein synthesis inhibitors.

Methods: MRC determinations followed Clinical Laboratory Standards Institute (CLSI) guidelines. Compounds were examined at 2× MIC for their inhibitory effects on the synthesis of RNA, DNA, and proteins in Escherichia coli ATCC 25922 as the test organism. The assay explored the incorporation of an appropriate radiolabeled precursor into a mass-produced subunit (53% for DNA, 49% for RNA, 10% for protein). Effects of compounds on various protein polymers was investigated by treating E. coli cells with different compound concentrations and analyzing polypeptide patterns by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE). Finally, compounds developed in this study were screened for their ability to inhibit protein synthesis in clinical isolates of E. coli (ATCC 25922) and of a human pathogens known to be resistant to known antibiotics by the disk diffusion method.

RESULTS AND DISCUSSION

An inhibitory concentration of 2× MIC was selected for determining the mechanism of action of E. coli ATCC 25922, a Gram-negative Escherichia coli (ATCC 25922) strain. The susceptibility of the 50S ribosomal subunit is critical to the success of protein synthesis inhibitors. The experiment was designed to examine the effects of the inhibitors on the inhibition of both RNA and DNA synthesis. The results showed that the compounds were able to inhibit protein synthesis in E. coli ATCC 25922, with an MIC of 6.25 µg/mL.

CONCLUSIONS

- The compounds displayed significant inhibition of protein synthesis in E. coli ATCC 25922.
- The compounds were effective against a wide range of Gram-negative pathogens.
- Further studies are needed to determine the clinical efficacy of these compounds.

REFERENCES