SYNTHESIS AND ANTIBACTERIAL ACTIVITY OF TRINEMS BEARING NITROGEN DERIVATIVES AT C(4)

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Optimization of the antibacterial activity of 4-amino trinem, obtained through chemical modification of the basicity of the amino group at position 4, has led to the identification of a very interesting compound characterized by a broad spectrum of activity including *Pseudomonas aeruginosa*.

β-Lactams [1-3] constitute a very important class of antibacterial agents which encompass penicillin, cephalosporins, monobactams, penems, carbapenems, and more recently trinems [4-11].

[Chemical structures of 1 and 2 are shown here.]

The general trend presented by 4-substituted trinems 1 is characterized by potency, broad spectrum of activity, remarkable resistance to β-lactamases, and stability to mammalian renal peptidases (DHP-1).

SAR studies [12, 13] have shown that the absolute stereochemistry at C(4) and C(8) of 4-substituted trinems is very important in modulating the antibacterial activity and other biological properties; in particular, (4S,8S) were recognized as the most effective absolute configurations.

Because of the well known epidemiological importance of *Pseudomonas aeruginosa* both in the hospital and community, the identification of a broad spectrum antibacterial compound covering this opportunistic pathogen would represent a very attractive area of research for many pharmaceutical companies involved in the antibacterial field.

Among other classes, 4-amino trinem derivatives have shown an interesting antimicrobial activity over a broad range of bacteria [14]; in particular, the activity against *P. aeruginosa* was comparable to imipenem (2), one of the best anti-infective agents currently in use. We thought that a compound encountering the aforementioned antibacterial profile would be obtained through a further derivatization of such a class.

As part of our research activity we focused our attention on the synthesis of a small set of trinem derivatives 3-6, characterized by the presence of a nitrogen at C(4), whose basicity was modulated by the introduction of an hydrophilic group.

[Chemical structures of 3 and 4 are shown here.]

SYNTHESIS

Trinems 3 [15, 16] and 4 [17] were obtained starting from the common and already described amino acid 7 [14], as outlined in Scheme 1.

Scheme 1

The reduced solubility of the amino acid 7 in many organic solvents forced us to perform the final conversion to 4-N-methylureido trinem 3 in water. Compound 7 was dissolved in water and treated with an excess of trimethylsilylisocyanate (TMSNCO) to give the crude 4-N-methylureido trinem 3, which was purified by reverse phase chromatography (RP-18). This compound quite rapidly underwent an intramolecular Michael addition giving the tetracyclic saturated compound 8. This irreversible reaction was followed by 1H NMR spectroscopy in D2O at 22 °C, obtaining an almost complete conversion to 8 in 24 h. The absolute stereochemistry of compound 8 was confirmed by NOE experiments and a more detailed analysis of such experiments will be reported elsewhere.

A pure sample of compound 8 when tested in vitro was proved to be completely inactive from the microbiological point of view.

A freshly purified sample of compound 3 was assayed against several bacterial strains (see Table 1), showing a significant antimicrobial activity including P. aeruginosa.

Compound 4 was prepared reacting benzyloxyformimidate hydrochloride with the amino acid 7 in a buffered solution at pH 8. The compound was purified by reverse phase chromatography and isolated after freeze drying and its microbiological profile evaluated.

The availability of 7 prompted us to attempt the synthesis of the other two 4-amino trinem derivatives 5 and 6 in the aqueous phase using the corresponding acylating agents, acetic anhydride and acetic formic anhydride (AFA) [12]; however, as expected, the reactivity of these systems in water was very poor giving the desired compounds in