N-(Trimethylsilyl)-2-Amino-5-Nitrothiazole; An Efficient Reagent for the Direct Synthesis of 2-Amino-5-Nitrothiazole-based Antimicrobial Agents

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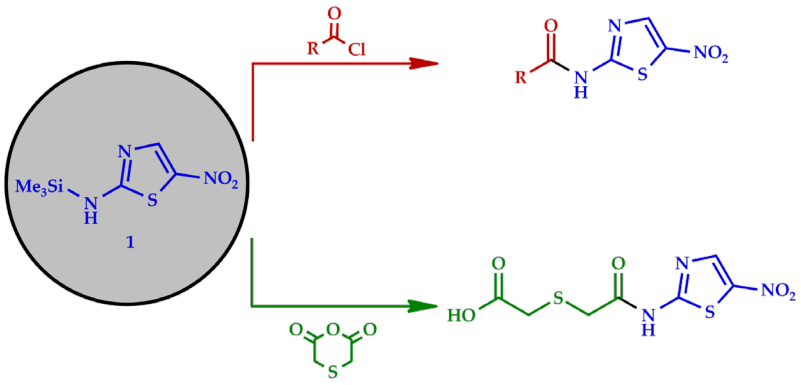

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Abstract Here we report the synthesis of a novel reagent designed to prepare 2-amino-5-nitrothiazole (ANT) amides and analogues in high yields. *N*-(Trimethylsilyl)-2-Amino-5-nitrothiazole [*N*-(TMS)-ANT] (**1**) was prepared in 99% yield via silylation of ANT (**2**) using 1,1,1,3,3,3-hexamethyldisilazane (HMDS), trimethylsilyl chloride (TMSCl) and catalytic saccharin. *N*-(TMS)-ANT (**1**) is a superb reagent for the preparation of ANT amides in excellent yields. Notably, cyclic anhydrides and base-sensitive acyl chlorides can be utilized with **1** to furnish ANT amides that are difficult to prepare by previously reported procedures.

Key words 2-amino-5-nitrothiazole; N-silylation; antimicrobials; antiviral; biofilms

Introduction

Nitazoxanide (**3**) (**Figure 1**), first synthesized by Rossignol and Cavier in 1975, is an orally bioavailable broad-spectrum antiparasitic and broad-spectrum antiviral drug that is the prototype for the thiazolide class of antimicrobials. It is a first-line treatment for Cryptosporidium parvum and Giardia lamblia infections. As an antiviral, it has shown efficacy for treating influenza, reducing shedding of the influenza virus, and holds promise as a broad-spectrum antiviral for respiratory infections[1]. The pharmacophore responsible for its biological activity is derived from 2-amino-5-nitrothiazole (**2**).

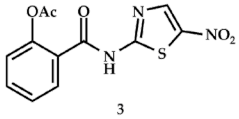
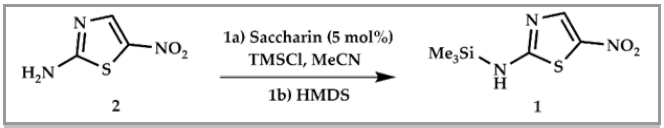


Figure 1: Nitazoxanide

In more recent years, ANT-based motifs have found applications in anti-biofilm research, showing promising activity against Escherichia coli, Staphylococcus epidermidis, Pseudomonas aeruginosa and Clostridium difficile[2-5]. Previously, most of the reported ANT-based amides were prepared via coupling of (**2**) with acyl chlorides in the presence of auxiliary bases. Standard procedures using ANT with Et_3N or K_2CO_3 as bases involve aqueous workups, which are capricious in that emulsions are often encountered, leading to variable yields (vide infra).[4,6] Subsequent purifications are also tedious, often requiring cautious column chromatography to obtain purified products. Since ANT derivatives have shown significant medicinal promise, an improved synthetic protocol for their synthesis was required.



Scheme 1: Synthesis of *N*-(Trimethylsilyl)-2-amino-5-nitrothiazole (**1**).

Here we report the synthesis of *N*-(Trimethylsilyl)-2-amino-5-nitrothiazole (**1**) [*N*-(TMS)-ANT], which efficiently couples with acyl chlorides, anhydrides, and other electrophiles directly in a 1:1 fashion. *N*-(TMS)-ANT (**1**) is efficiently prepared via silylation of ANT (**2**) using 1,1,1,3,3,3-hexamethyldisilazane (HMDS), trimethylsilyl chloride (TMSCl) and catalytic saccharin (**Scheme 1**).[7]

The HMDS derived byproduct NH_3 was found to be deleterious to the course of this reaction, so it was scavenged by the preliminary addition of TMSCl. Using this procedure, the title compound (**1**) was obtained in near quantitative (99%) yield.

Results and Discussion

The purification of commercial ANT was found crucial to the preparation of *N*-(TMS)-ANT (**1**) in high yield. Previously this was accomplished by recrystallization of ANT from large quantities of ethanol, which we found to be impractical, since

dark impurities were difficult to remove using this solvent. Accordingly, we developed an alternative purification method involving recrystallization from CH_3CN. Importantly, in the preparation of *N*-(TMS)-ANT (**1**), it was found critical to ensure that all reagents were anhydrous, since the presence of water led to incomplete conversion.

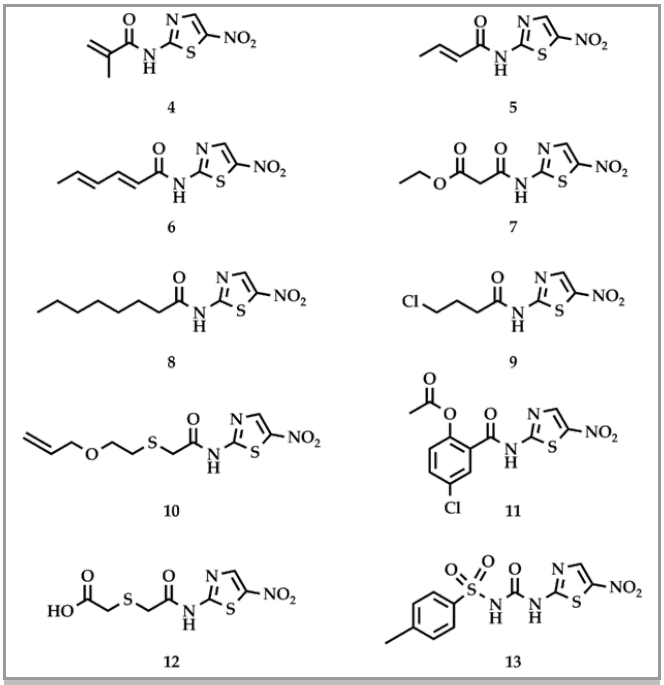


Figure 2: ANT derivatives prepared via coupling with N-(TMS)-ANT (**1**).

N-(TMS)-ANT (**1**) was subsequently treated with various electrophiles, to provide the corresponding derivatives shown in **Figure 2**. The utilization of *N*-(TMS)-ANT (**1**) markedly improved the preparative efficiency leading to five previously reported antimicrobial agents (**Figure 1**; 4, 8, 9, 11, 13) from 87, 38, 86, 74, and 22% to enhanced yields of 89, 86, 97, 98, and 93% respectively (**Table 1**).

Table 1: Yields of coupling reactions using N-(TMS)-ANT (**1**) compared to those previously cited in the literature.

Compound	Yield (%)	Literature Yield (%)
4[a,c]	89	87 [4]
5[a]	95	
6[a]	85	
7[a,b]	74	
8[a]	86	38 [6]
9[a]	97	86 [5]
10[a,c]	79	
11[a]	98	74 [4]
12[a]	73	
13[a]	93	22 [8]

[a]Compounds selected based on functionality that prior SAR studies have shown to be important. [b]Extended *n*-alkyl and ester moieties are of interest (e.g., **7**) in anti-biofilm applications. [c]Terminal alkenes (e.g., **4** and **10**) are also of interest, as they present the opportunity for union with larger molecular motifs via alkene cross-metathesis.

Additional couplings using carboxylic anhydrides and a *N*-sulfonyl isocyanate expanded the library of available ANT compounds as well as the scope of electrophilic functionalization. It is of particular significance in a synthetic context that acyl chlorides **5** and **6** undergo preliminary deconjugative deprotonation in the presence of Et_3N, leading to acyl-rearranged product mixtures. In addition, the use of Et_3N with the labile acyl chloride **7** under standard basic conditions resulted in its premature decomposition. Moreover, the reaction of cyclic anhydrides and sulfonyl isocyanates (corresponding to **12** and **13**) with **1** directly provides the corresponding TMS intermediates, which are converted to the desired products by simple passage through a pad of silica gel, thereby obviating inadequate aqueous-reliant purification methods.

Conclusion

N-(TMS)-ANT (**1**), a new reagent for the efficient, high-yield synthesis of ANT derivatives has been efficiently prepared in excellent yield. Primary advantages associated with the use of **1** include its ability to cleanly amidate base-labile acyl chlorides as well as cyclic anhydrides. Additional applications of **1** for the synthesis of other types of ANT-containing antimicrobials will be the topic of future reports from this laboratory. Future studies will also include the biological evaluation of novel ANT-based antimicrobials for the control of biofilm-forming bacteria.

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Supporting Information

YES (this text will be updated with links prior to publication)

Primary Data

NO.

Conflict of Interest

The authors declare no conflict of interest.

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(9) **Preparation of *N*-(TMS)-ANT** (**1**): A 100 mL round-bottomed flask equipped with a magnetic stirring bar and a poly-seal cap was charged with 2-amino-5-nitrothiazole (3.625 g, 25.0 mmol), saccharin (0.169 g, 1.25 mmol) and anhydrous acetonitrile (25 mL). The reactant mixture was blanketed with argon and then trimethylsilyl chloride (1.05 g, 1.225 mL, 9.5 mmol) was added dropwise by syringe with stirring. After stirring for an additional 10 min at 23 ˚C, 1,1,1,3,3,3-hexamethyldisilazane (1.48 g, 1.91 mL, 9.17 mmol) was added dropwise by syringe. The reactant mixture was stirred for 1 h at 23 ˚C and then heated at 65 ˚C with stirring

for an additional 1 h. The NH_4Cl precipitate was then filtered, with care being taken to avoid exposure to atmospheric moisture. The NH_4Cl was leached with anhydrous acetonitrile (5 mL) and the solvents were removed in vacuo. A 100 mL recovery flask was used for the final removal of trace solvents and subsequent sublimation under high vacuum (0.01 mmHg 60 ˚C) furnished the title compound **1** as a yellow/orange crystalline solid (5.4 g, 99%). **^{1}H-NMR** (CDCl3, 500 MHz): δ 8.08 (1H, s), 5.30 (1H, s), 0.39 (9H, s). **^{13}C-NMR** (CDCl3, 125 MHz) δ 169.7, 143.1, 29.9, 0.0. **HRMS** Electrospray ionization (ESI) m/z calcd. for [C6H11N3O2SSi + H]+ : 218.0414, found: 218.0425 (mass error Δm = 5 ppm).

(10) **Representative Amidation; Preparation of 5:** A 10 mL round-bottomed flask equipped with a magnetic stirring bar and poly seal cap was charged with *N*-(TMS)-ANT (**1**) (217 mg, 1.0 mmol), anhydrous dichloromethane (3.0 mL), and blanketed with argon in succession. The mixture was stirred until homogeneous, then crotonoyl chloride (105 mg, 96 µL, 1.0 mmol) was subsequently added by gas-tight syringe and the reactant mixture was allowed to stir at room temperature for 24 h. The solvent was then removed in vacuo and the residue was diluted with EtOAc (5 mL) and successively extracted with water (2 × 5 mL), saturated sodium bicarbonate (2 × 5 mL), and brine (2 × 5 mL). The organic layer was then passed through a pad of silica gel and removal of the solvent in vacuo provided the title compound (202 mg, 95%). **^{1}H-NMR** (D_6-DMSO, 500 MHz): δ 13.13 (1H, s), 8.65 (1H, s), 7.10 (1H, dq, *J* = 7 Hz), 6.26 (1H, dq, *J* = 7, 2 Hz), 1.94 (3H, dd, *J* = 7, 2 Hz). **^{13}C-NMR** (D_6-DMSO, 125 MHz) δ 164.8, 162.5, 146.6, 143.3, 142.5, 123.0, 18.6. **HRMS** Electrospray ionization (ESI) m/z calcd. for $[C_7H_7N_3O_3S]^+$: 213.0208, found: 213.0146 (mass error Δm = 29 ppm).