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Serendipitous Discovery of a Zidovudine Guanidine Complex: A Superior Process for the Production of Zidovudine

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Supporting Information

ABSTRACT: A superior process for the commercial production of zidovudine (AZT) has been developed. It was discovered that an AZT−guanidine complex formed when a crude zidovudine solution was treated with guanidine. This readily precipitated from protic solvents resulting in the exclusion of impurities and permitted the development of a superior isolation and purification of AZT.

INTRODUCTION

Zidovudine (AZT), first synthesized by Horwitz et al.[1] in 1964, was the first antiviral agent approved for the clinical treatment of HIV infections.[2] In their procedure, thymidine was tritylated,[3,4] mesylated,[4] and reacted with excess sodium hydroxide to give, directly or via the corresponding 2,3′-anhydro intermediate (TCT **2**), 5′-*O*-trityl-*threo*-thymidine **5**,[4−6] which was mesylated,[1] reacted with lithium azide,[1] and deprotected[1] with hydrochloric acid to give AZT **4** in an overall yield of 35.7%[1] (Scheme 1). Glinski et al.[7] reported a similar but shorter procedure (Scheme 1) using sodium azide and 5′-*O*-trityl-2, 3′-anhydrothymidine (TCT **2**), to give 3′-azido-3′-deoxy-5′-*O*-tritylthymidine (Trityl-AZT **3**) which was deprotected with acetic acid to AZT **4** in an overall yield of 17−18% from thymidine. Although the yield was only about one-half of Horwitz's process, it was more efficient in terms of unit process operations, chemical steps and the amounts of reagents used.

Our department was charged with designing a superior synthesis of AZT with the object of eventually establishing a lower-cost large-scale manufacture of this important drug. After reviewing a number of other procedures,[8] I decided to follow the approach of Glinski[7] in starting with TCT **2** for the introduction of the azido group.

RESULTS AND DISCUSSION

The preparation of TCT **2** was improved to an average yield of 85% starting from thymidine by an approach which was similar to the one developed by Horwitz.[5] The azidation reaction was modeled on Glinski's approach in that the starting material was TCT **2**, but the solvent was changed to dimethyl sulfoxide (DMSO) because DMF gave blackish solutions and the resultant AZT **4** was difficult to crystallize. Initially the amount of sodium azide was reduced from 3.7 equiv to 1.5, but after an impurity, which is discussed below, had been identified, the amount was increased to 2.5 equiv, and the impurity was reduced from ∼7% to ∼4%, the deprotection was done with aq HCl in methylisobutyl ketone (MIBK) instead of 80% acetic acid, and the AZT **4** was separated from the trityl alcohol by rendering the mixture alkaline, driving sodium AZT **4** into the aqueous phase. The alkaline phase was neutralized, extracted with MIBK, and crystallized to give crude AZT **4** in an initial average yield of 67% and eventually optimized to a yield of 81%. The crude AZT **4** was recrystallized from aq acetone to >99.5% purity AZT **4** eventually in 73% optimized recovery. The final optimized yield for pure AZT **4** from thymidine was 50%.

The process was efficient from an operational point of view but it produced 3−7% of a major dimer side product **7** [3′-*N*″-(3‴-azido-3‴-deoxythymid-3″-yl)-3′-deoxythymidine] (Figure 1).[9a,b] Two other major side products were also produced: thymine **8** [3−7%] which is a degradation product of the thymidine derivatives present in the reaction mixture and 3-*N*-AZT **9** [2−5%, 3-(3′-azido-2′,3′-dideoxy-β-D-*erythro*-pentofuranosyl)thymine] which is a degeneration and regeneration product at the N3 from trityl AZT **3**,[10] however **7** was the most difficult to separate, requiring large volumes of acetone during the final recrystallization step. This also impacted the % recovery.

To better understand the formation of this major impurity, the dimer **7** was isolated by chromatography, and the structure was confirmed by ^{1}H NMR and ^{13}C NMR spectral analysis, elemental analysis, IR, UV and mass spectrometry. The proposed mechanism for the formation of the **7** is shown in Scheme 2. Also the knowledge of knowing the structure of **7** allowed for an early improvement in its reduction from ∼7% to ∼4% by increasing the amount of sodium azide from 1.5 to 2.5 equiv.

TCT **2** reacts with sodium azide which may give the salt sodium trityl AZT **10**, which then attacks another molecule of TCT **2** to furnish the salt sodium ditrityl dimer **11** which upon deprotection gives **7**. In Scheme 2 the sodium salts **10** and **11** are shown with the negative charges on the C2-oxygens rather than nitrogen because oxygen is more electronegative than nitrogen. It is also possible to put the negative charge on the C4-oxygen, but the ^{13}C NMR chemical shifts of sodium trityl AZT **10** for C2 at δ 157.84 ppm and C4 at δ 173.79 ppm fit very closely to the simulated spectral[11] values at 159.62 and 173.75, respectively, when the negative charge is placed on the C2-oxygen.

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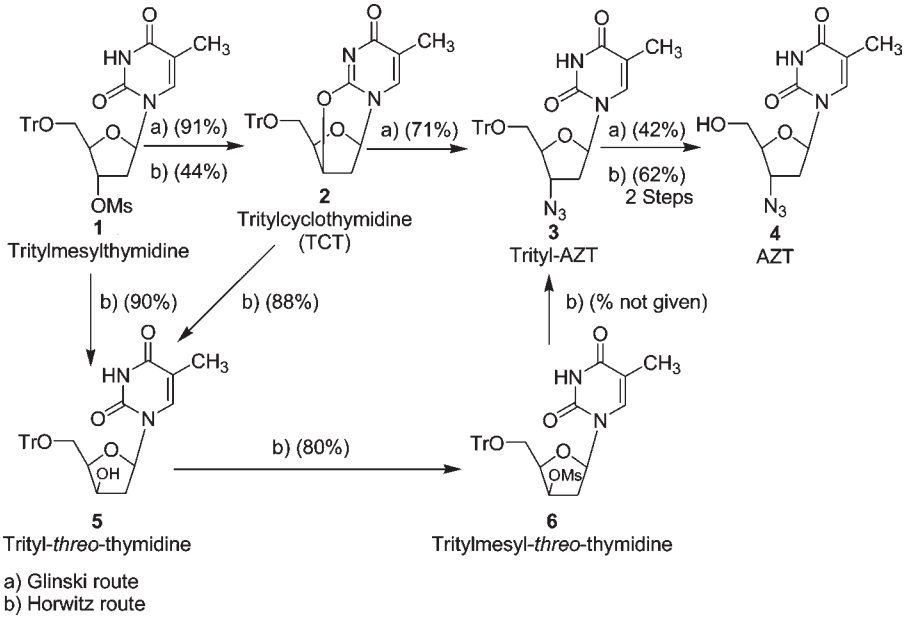
Scheme 1. Processes by Glinski (a) and Horwitz (b) for the preparation AZT 4



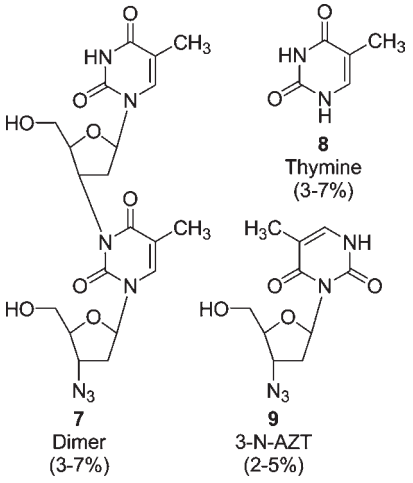


Figure 1. Major impurities in the production of AZT **4**.

The corresponding simulated values for when the negative charge is placed on the C4-oxygen and N3 are 161.99 and 177.60 and 163.00 and 180.00, respectively. Chemical support for this mechanism comes from the reaction between pure trityl AZT **3** and TCT **2** in DMSO in the presence of potassium carbonate at 108 °C for 24 h. The reaction mixture was worked up, treated with acid, and worked up to a syrup. HPLC of the syrup indicated **7** (27 area %), thymine (16%), and AZT **4** (31%). The syrup was purified to furnish **7** in 8.6% yield.

The separation of **7** from AZT **4** could be achieved by crystallization using a number of different solvent combinations but with a significant loss of yield. A number of process variations before crystallization were tried, such as acid/base liquid/liquid extractions that also proved to be unsuccessful in removing this impurity. In a search to find means to manage the problem posed by this side product in the reaction and its propensity to remain with the product AZT **4**, the focus was shifted to finding a way to modify **7**, thereby forming products which may be more readily separated from crude AZT **4**. A number of ultimately unsuccessful approaches were studied, including degradation by either acid or base, and complexation or salt formation. Derivatization was not considered since this would entail at least one additional chemical step. However, the literature[12,13] suggested a different approach in that of uracil, 1-methyluracil, 3-methyluracil and 1,3-dimethyluracil, only 1,3-dimethyluracil would react with guanidine (Scheme 3 based on description given in reference[12]).

The explanation for the nonreactivity of the other three compounds (see ref 12) was that in the presence of a strong base such as guanidine the presence of acidic protons would deprotonate the bases, rendering nucleophilic attack more difficult[12] or, in other words, an electron-rich nucleophile such as guanidine would be repulsed by an electron-rich pyrimidine anion. An examination of the structures of AZT **4** and **7** revealed that, for the former, the base moiety was monoalkylated and would be predicted, according to the above explanation, not to be susceptible to attack by guanidine because the nucleo-base would become anionic; for **7** the middle base unit was dialkylated and thus would be predicted to be susceptible to attack by guanidine because the nucleo-base would remain neutral. A possible outcome is shown in Scheme 4. The resulting urea or further degradation products may be more easily removed from the product mixture than **7**. This analysis provided sufficient justification for us to examine this approach.

Some dimer **7** was combined with a methanolic solution of guanidine, evaporated to a thin syrup, and heated. After 2.5 h, TLC showed only a trace of **7**. This result seemed to support the proposed mechanism of Scheme 4. In another experiment, AZT **4** syrup containing 83.2 HPLC area % of AZT and 9.38% of **7**, was dissolved in ethanolic guanidine at reflux. After 29 h, it showed little if any change by TLC. However, when the mixture was evaporated to give a syrup, heated at 80 °C for 6.5 h to replicate fusion conditions, and allowed to cool, it had solidified by the next day. In spite of further manipulation by adding more

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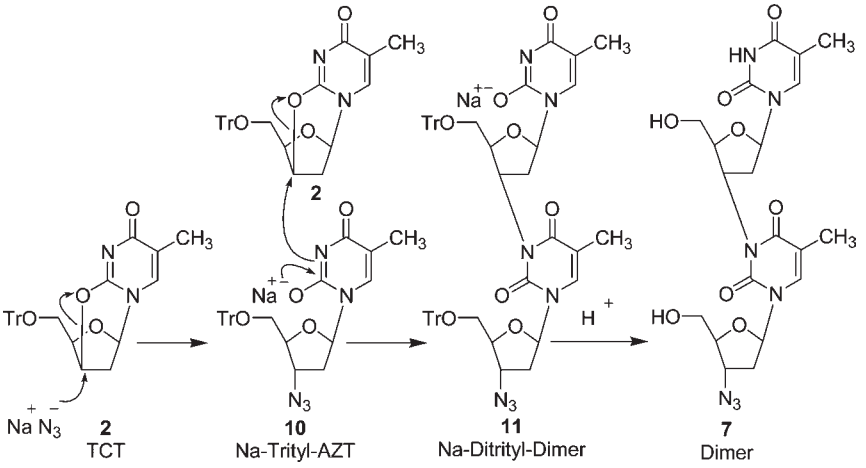
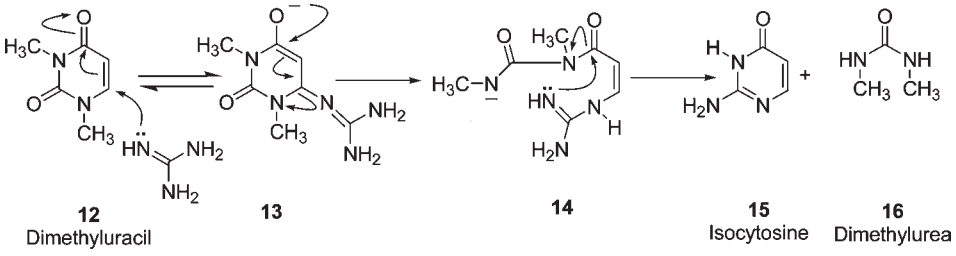
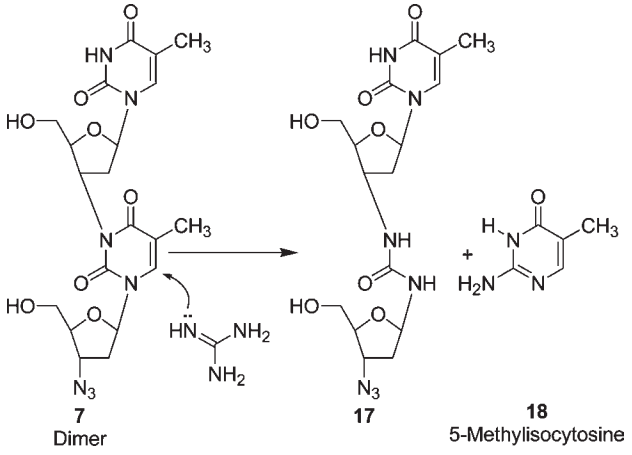
Scheme 2. Possible mechanism for the formation of dimer 7 during AZT 4 production



Scheme 3. Mechanism for the modification of dimethyl uracil upon treatment with guanidine



Scheme 4. Expected outcome of the treatment of dimer 7 with guanidine



guanidine and exchanging the solvent to other alcohols such as methanol or isopropanol, the solid could not be dissolved. Interestingly, much solid still remained even at reflux, an observation which was surprising since it was known that the solvents should have dissolved all of the available AZT **4** at reflux. A white solid was obtained after cooling and washing with isopropanol whose ^{1}H NMR spectrum showed the characteristic AZT **4** peaks, although slightly shifted, and a broad exchangeable singlet integrating for seven protons between the H-1′ and H-6 signals of AZT **4**. I believed that AZT **4** had formed a salt or complex with guanidine that, unlike the salt AZT sodium **19**, precipitated readily from the solvent. If AZT guanidine **20** is a salt, then the question arises as to where the negative charge should is located. The negative charge could be on the C2−O or N3 or C4−O, and C2−O is chosen for the same reasons given earlier for the case of the ^{13}C NMR downfield shifts of the C2 and C4 resonance when going from trityl AZT **3** to sodium trityl AZT **10**. The C2 and C4 resonances of 150.42 and 163.73, respectively, for AZT **4** shift downfield to 157.86 and 173.96, respectively, for AZT guanidine **20** which is in line with the simulated spectra[11] where the corresponding values are 150.33 and 163.33 for AZT **4** going to 159.62 and 173.75 ppm for AZT guanidine **20**. Also the actual ^{13}C NMR of sodium AZT **19** shows downfield shifts to 157.82 and 173.68 ppm, respectively, which are very close to the actual shifts for AZT guanidine **20**. The discovery of this AZT guanidine salt **20** was very exciting and changed the course of the study from base degradation to salt formation.

These initial studies demonstrated that the content of **7** could be lowered from 19% to 7% along with a 75% recovery of AZT. However, I wished to determine if such purification could be achieved at an earlier stage in the process. An attractive point would be when AZT **4** had been separated from the trityl alcohol by the water/MIBK extraction, which would avoid the additional steps of neutralization, extraction, and concentration. Solid guanidine hydrochloride was added to basic (pH ∼12) AZT **4** in aqueous solution, and the pH was adjusted to 12.2−12.3 with

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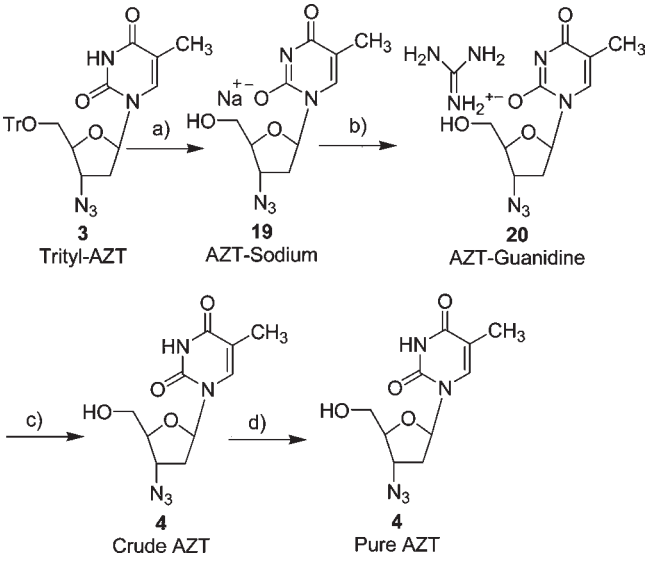
Scheme 5. Improved process for the production of AZT 4[a]



[a] Reagents and conditions: a) MIBK, 77 °C, 23% aq HCl, then RT, aq NaOH, pH 12, separate; b) aqueous phase, 70 °C, solid guanidine HCl, aq NaOH, pH 12, cool, filter 78−82%; c) H_2O, HCl, or AcOH, 75 °C, then **20**, cool, filter, 81−85%; d) H_2O, acetone, carbon, filter, partly distill acetone at atm, 35 °C, seeds, 23 °C, 12 h, vacuum distill acetone, 5 °C, filter, dry, 88−92%.

50% NaOH. AZT guanidine salt **20** readily precipitated from solution with a dimer **7** content of usually <1%. The modification achieved the desired purity result, eliminating three steps and avoided the need to separately prepare guanidine in alcohol from guanidine hydrochloride even though it introduced an additional step of releasing AZT **4** from the salt.

Releasing the AZT **4** from the guanidine salt was achieved by adding the salt to hot water containing one equivalent of hydrochloric acid or acetic acid, whereupon the precipitate dissolved and then another precipitate formed which was AZT **4** largely free of guanidine and dimer **7** (typically <1%). The resulting crude AZT **4** could then be purified by recrystallization in ∼90% recovery from water/acetone as previously described with an additional operation of vacuum distilling most of the acetone. This sequence of precipitating the AZT guanidine salt **20** was incorporated into the existing process and became the new commercial process (Scheme 5).[9a]

This new technology ultimately had a number of advantages over that of the original process. First, the AZT **4** is initially separated from the reaction mixture as AZT-guanidine **20** by precipitation, thus avoiding the tedious, highly dilute, and time-consuming liquid/liquid extraction, followed by concentration to obtain crystals. Second, the resultant AZT-guanidine **20** had a low dimer **7** content relative to the original formulation of this process which also led to crude AZT **4** of low dimer **7** content. Third, the purer crude AZT **4** allowed a more facile removal of the acetone during the final purification step leading to an improved recovery. The fourth advantage was that the water-damp AZT guanidine **20** salt and the crude crystalline AZT **4** did not have to be dried before going to the next step. Finally, the facile precipitation of AZT guanidine **20** allowed the recovery of a second crop of AZT **4** from the mother liquors in spite of the high levels of dimer **7** present. Eventually, once the process had been optimized, the yield of the first crop of pure AZT **4** was ∼55%, and the second crop increased the overall yield to ∼62%.

CONCLUSION

The commercial process for the production of AZT **4** has been improved by the discovery that AZT **4** forms a salt with guanidine which precipitates from aqueous solutions quite readily while an otherwise difficult-to-remove dimer remained in solution. The AZT guanidine salt **20** could be easily converted to crude AZT **4** in high recovery. This development significantly improves the purification of AZT and is currently used in our company's commercial production of AZT.

EXPERIMENTAL SECTION

The Initial Commercial Process for the Production of 3′-Azido-3′-deoxythymidine (AZT) 4. *Step A: Preparation of Crude AZT* ***4***. Tritylcyclothymidine (TCT) **2** (163 kg, 349.4 mol) was reacted with sodium azide (56 kg, 862.5 mol, 2.46 equiv) in DMSO (410 kg) at ∼110 °C for 24 h and was cooled to about 60 °C; MIBK (647 kg) preheated to ∼60 °C was added, followed by brine (195 kg) preheated to ∼60 °C and water (195 kg) preheated to ∼60 °C (preheating prevented the formation of taffy-like yellow globs); the phases were separated. Water (130 kg) and brine (130 kg) were added to the MIBK phase, cooled to about 35 °C, and separated. The combined aqueous phases were further extracted with MIBK (2 × 107 kg), and the combined MIBK phases were washed once with brine (195 kg) to give trityl AZT **3** as an MIBK solution (1230 L). The MIBK solution was heated to 77 °C, and 23% hydrochloric acid (56.4 kg) was added. The reaction was monitored by TLC (ethyl acetate/hexane, 3:2), and after 1.5−2.5 h there was only a trace of trityl AZT **3** remaining. Heating was stopped, water (225 kg) was added, and the pH was adjusted to 12.05 with 50% NaOH (∼40 kg). The phases were separated, and the MIBK phase was extracted twice with water (64 and 33 kg). The combined aqueous phases were diluted with MIBK (445 kg), and the pH was adjusted to 7.02 with 16% hydrochloric acid (87.6 kg). The phases were separated, and the aqueous phase was vacuum-distilled until 330 L of distillate was collected and extracted with MIBK (multiple times with a total of 330 kg). Celite (4.7 kg) was added to the combined MIBK phases and filtered. The MIBK filtrate was vacuum concentrated until about 765 L of distillate was collected, cooled to 25 °C, seeded, and maintained for 6 h. The mixture was further cooled to 5 °C and maintained for 24 h. The crystals were filtered, washed with MIBK (65 kg), and dried to give 78.6 kg (294.1 mol, 84.2%) of crude AZT **4**, (HPLC purity = 94.3 area %, dimer **7** content = 4.5%).

Step B: Preparation of Pure AZT ***4***. Crude AZT **4** (290 kg, HPLC purity = 93.8%, dimer = 4.7%) was dissolved in a mixture of water (870 kg) and acetone (540 kg) at 50 °C. Active carbon (7.25 kg) and Celite (20 kg) were added, the mixture was filtered over a Celite bed at 30 °C, and the bed was washed with additional acetone (3 × 45 kg). Acetone (∼380 L) was distilled at atmospheric pressure, and a sample was checked by NMR to determine the water/acetone ratio which was 2.2 and which was in the acceptable range (2.1−2.5). The mixture was cooled to 35 °C, seeded with pure AZT **4**, and agitated for 12 h. The mixture was further cooled to 4 °C and maintained for 1 h; a sample was filtered and checked by HPLC for the dimer content

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which was 0.15% (required end point was <0.2%). The mixture was filtered, washed with water (3 × 35 kg), vacuum-distilled (25−30 in.), and dried at 55−65 °C for 12−14 h to yield 210.7 kg of AZT **4**: HPLC purity = 99.7%, dimer **7** content = 0.15%. Calcd for $C_{10}H_{13}N_5O_4$: C, 44.94; H, 4.90; N, 26.21. Found: C, 44.85; H, 4.87; N, 26.15. UV: λ_{max} (H_2O) 208, 268 nm. IR (KBr disk): 2115 cm^{-1} (azide). MS (EI) *m/z*, 267 (M^+), 142 (sugar moiety), 126 (thymine). ^{1}H NMR (300 MHz DMSO-d_6) δ 1.78 (s, 3H, CH_3), 2.23−2.43 (m, 2H, H-2′s), 3.62 (m, 2H, H-5′s), 3.82 (dd, 1H, *J* = 4.0, 8.5 Hz, H-4′), 4.41 (dd, 1H, *J* = 5.2, 12.0 Hz, H-3′), 5.23 (bs, 1H, exchangeable, OH), 6.10 (t, 1H, *J* = 6.4 Hz, H-1′), 7.69 (s, 1H, H-6), 11.33 (bs, 1H, NH). ^{13}C NMR (75 MHz DMSO-d_6) δ 12.24 (CH3), 36.22 (C-2′), 60.19 (C-3′), 60.83 (C-5′), 83.43 (C-1′), 84.01 (C-4′), 109.54 (C-5), 136.06 (C-6), 150.42 (C-2), 163.73 (C-4).

Isolation of 3′-*N*″-(3‴-Azido-3‴-deoxythymid-3″-yl)-3′-deoxythymidine (7). The MIBK mother liquor produced from processing 13.3 kg of tritylcyclothymidine was concentrated to 2.5 L, cooled, seeded, and held for 10 d. The crystals were filtered, washed with MIBK, and dried to give about 600 g of crystals containing ∼15% **7**. The crystals were dissolved in ethyl acetate (1 L) and applied to a silica gel column (1 kg) and eluted with ethyl acetate/hexanes (7:3). Appropriate fractions were combined and recrystallized from ethanol to give about 40 g of **7**. Mp 215.2−216.7 °C. Anal. Calcd for $C_{20}H_{25}N_7O_8$: C, 48.86; H, 5.13; N, 19.96. Found: C, 48.54; H, 5.06; N, 19.75. UV: λ_{max} (H_2O) 207, 268 nm. IR (KBr disk): 2101 cm^{-1} (azide). MS (ESI$^+$) *m/z*, 514.19 (67.8%)(M + Na^+). ^{1}H NMR (500 MHz DMSO-d_6) δ 1.78 (s, 3H, CH3″), 1.84 (s, 3H, CH_3), 2.15 (m, 1H, H-2_1′), 2.33 (m, 1H, H-2_1‴), 2.43 (m, 2H, H-2_2′, H-2_2‴), 3.49 (m, 1H, H-5_1′), 3.56 (m, 1H, H-5_2′), 3.59 (m, 1H, H-5_1‴), 3.67 (m, 1H, H-5_2‴), 3.83 (m, 1H, H-4‴), 4.11 (m, 1H, H-4′), 4.38 (m, 1H, H-3‴), 4.97 (t, 1H, exchangeable, 5′-OH), 5.23 (t, 1H, exchangeable, 5‴-OH), 5.57 (m, 1H, H-3′), 6.14 (t, 1H, H-1‴), 6.56 (t, 1H, H-1′), 7.78 (s, 2H, H-6, and H-6″), 11.27 (s, 1H, exchangeable, NH). The ^{1}H NMR assignments were made by comparison with the spectrum for 3′-azido-3′-deoxythymidine **4** and by analysis of a two-dimensional (COSY) experiment which allowed the assignment of the sugar protons to the appropriate furanose ring. ^{13}C NMR (75 MHz DMSO-d_6) δ 12.20 and 12.97 (CH_3 and CH_3″), 34.84 and 36.57 (C-2′ and -2‴), 50.76 and 59.68 (C-3′ and -3‴) 60.55 and 62.03 (C-5′ and -5‴), 81.71 and 84.23 (C-4′ and -4‴), 84.45 and 84.77 (H-1′ and -1‴), 108.82 and 109.66 (H-5 and -5″), 134.92 and 136.78 (C-6 and -6″), 150.30 and 150.51 (C-2 and -2″), 162.87 and 163.75 (C-4 and -4″).

Synthesis of 3′-*N*″-(3‴-Azido-3‴-deoxythymid-3″-yl)-3′-deoxythymidine (7). Trityl AZT **3** (5.1 g, 0.01 mol, prepared from pure AZT **4** to ensure no contamination with dimer **7**) was combined with TCT **2** (9.33 g, 0.02 mol, 2 equiv) in DMSO (25 mL) and heated to dissolve the solids. Potassium carbonate (2.8 g, 2 equiv) was added to the mixture, heated to 108 °C, and maintained for 24 h. The mixture was diluted with MIBK (75 mL), brine (50 mL) and water (50 mL), the pH was adjusted to about 7 with aq HCl and the phases were separated. The aqueous phase was extracted with MIBK (3 × 50 mL). The combined MIBK phases were vacuum evaporated on a rotary evaporator to a solid foam (14.78 g). This was dissolved in ethanol (50 mL), chloroform (25 mL), and trifluoroacetic acid (25 mL). After 75 min at ambient temperature, TLC showed that the reaction was complete, and the reaction mixture was vacuum evaporated (rotary evaporator) at ∼40 °C. Methanol (3 × 50 mL) was added and evaporated three times. The syrup was dissolved in acetone (15 mL) and water (100 mL) and agitated overnight. The trityl alcohol was filtered and washed with water (15 mL); the aqueous filtrate was vacuum evaporated to a syrup (4.45 g). The syrup was preabsorbed on silica gel (10 g), applied to a silica column (60 g), and eluted with ethyl acetate. Appropriate fractions were combined and evaporated, and the residue was crystallized from ethanol to give 0.42 g (8.6%) of dimer **7**.

Preparation of Trityl AZT 3 and Sodium Trityl AZT 10 for Use in NMR Comparisons. The trityl AZT **3** which was used to obtain NMR data and to convert to sodium trityl AZT **10** was prepared from AZT **4** (26.7 g) and trityl chloride (35.4 g) in pyridine (100 mL) heated at 70−75 °C for 1 h and then kept at room temperature for 20 h. A standard workup gave a syrup which was purified by column chromatography to yield 41.18 g of trityl AZT **3** as a white foam. ^{1}H NMR (400 MHz DMSO-d_6) δ 1.57 (s, 3H, CH_3), 2.37 and 2.50 (2*m*, 2H, 2 × H-2′), 3.27 (m, 2H, H-5′s), 3.89 (m, 1H, H-4′), 4.60 (dd, 1H, *J* = 6.4, 13.3 Hz, H-3′), 6.15 (t, 1H, *J* = 6.4 Hz, H-1′), 7.26−7.42 (m, 15H, trityl), 7.54 (s, 1H, H-6), 11.39 (s, 1H, NH). ^{13}C NMR (100 MHz DMSO-d_6) δ 11.86 (CH_3), 36.98 (C-2′), 59.72 (C-3′), 63.11 (C-5′), 81.94 (C-1′), 83.31 (C-4′), 86.49 ($Ph_3\underline{C}$), 109.73 (C-5), 127.19, 127.97, 128.23, and 143.35 ($\underline{Ph_3}C$), 135.85 (C-6), 150.35 (C-2), 163.64 (C-4).

Trityl AZT **3** (0.509 g) and 50% sodium hydroxide (0.080 g, 1 equiv) were dissolved in methanol (20 mL) and evaporated in vacuo to a solid foam to give sodium trityl AZT **10**. ^{1}H NMR (400 MHz DMSO-d_6) δ 1.51 (s, 3H, CH_3), 2.32 and 2.50 (2 × m, 2H, 2 × H-2′), 3.23 (m, 2H, H-5′s), 3.82 (m, 1H, H-4′), 4.49 (m, 1H, H-3′), 6.29 (t, 1H, *J* = 6.8 Hz, H-1′), 7.21−7.40 (m, 15H, trityl), 7.43 (s, 1H, H-6). ^{13}C NMR 100 MHz DMSO-d_6) δ 13.46 (CH_3), 36.32 (C-2′), 60.50 (C-3′), 63.55 (C-5′), 81.22 (C-1′), 83.11 (C-4′), 86.45 ($Ph_3\underline{C}$), 109.77 (C-5), 127.15, 127.96, 128.26, and 143.45 ($\underline{Ph_3}C$), 133.32 (C-6), 157.84 (C-2), 173.79 (C-4).

Preparation of Sodium AZT 19 for Use in NMR Comparisons. AZT **4** (0.267 g) and 50% sodium hydroxide (0.080 g, 1 equiv) were dissolved in methanol (20 mL) and evaporated in vacuo to a solid foam to give sodium AZT **19**. ^{1}H NMR (400 MHz DMSO-d_6) δ 1.57 (s, 3H, CH_3), 2.37 and 2.50 (2*m*, 2H, 2 × H-2′), 3.27 (m, 2H, H-5′s), 3.82 (dd, 1H, *J* = 4.0, 8.5 Hz, H-4′), 4.41 (dd, 1H, *J* = 5.2, 12.0 Hz, H-3′), 5.23 (bs, 1H, exchangeable, OH), 6.10 (t, 1H, *J* = 6.4 Hz, H-1′), 7.69 (s, 1H, H-6), 11.33 (bs, 1H, NH). ^{13}C NMR (100 MHz DMSO-d_6) δ 13.69 (CH_3), 36.08 (C-2′), 61.03 (C-3′), 61.34 (C-5′), 83.35 (C-1′), 83.68 (C-4′), 109.57 (C-5), 134.04 (C-6), 157.82 (C-2), 173.68 (C-4).

New Commercial Process for the Production of 3′-Azido-3′-deoxythymidine (AZT) 4. *Step A: Preparation of AZT Guanidine Salt **20**.* Tritylcyclothymidine (TCT) **2** (250 kg, 535.9 mol) was reacted with azide and further processed as described in Step A of the first example of the Experimental Section to give 900 L of a solution (pH 12−13) containing sodium AZT **19**. The solution was heated to 70 °C, solid guanidine hydrochloride (61.3 kg, 641.9 mol, 1.2 equiv) was added, a precipitate formed and the pH was adjusted to 12.2−12.3 with 50% NaOH (∼6 kg). The mixture was vacuum-distilled (350−550 mmHg) at ∼60 °C to a volume of about 300 L to give a pastelike mixture, to which was added methanol (200 kg). The mixture was heated to reflux, maintained for 15 min (crystals remained), cooled to 5 °C, maintained for 6 h, filtered, and washed with water (2 × 50 kg) to give 186 kg of water-damp AZT guanidine **20**. A loss on drying determination

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(LOD = 24.1%) gave a dry weight of 141.2 kg (433.1 mol, 80.8%). An HPLC analysis: 94.54%/0.34% for AZT **4**/dimer **7**.

An analytical sample of 3′-azido-3′-deoxythymidine guanidine salt **20** was prepared by suspending clean 3′azido-3′-deoxythymidine **4** (53.4 g. 0.2 mol) in water (250 mL) and adding 50% NaOH (17.6 g, 0.22 mol) to pH 13.0. The mixture was heated to 70 °C. Thereafter, guanidine hydrochloride (23 g, 0.24 mol) was added and heated to 97 °C to dissolve the emerging precipitate. The mixture was set aside to cool with agitation to crystallize for 16 h. The crystals were filtered, washed with water (50 mL) and ethanol (50 mL), and dried to 58.76 g (0.18 mol, 90%) of white crystals: mp 210.9−212 °C. Anal. Calcd for $C_{11}H_{18}N_8O_4$: C, 40.49; H, 5.56; N, 34.34. Found: C, 40.92; H, 5.45; N, 34.17.UV: λ_{max} (H_2O) 268 nm. IR (KBr disk): 2095 cm^{-1} (azide). ^{1}HNMR (500 MHz DMSO-d_6) δ 1.71 (s, 3H, CH_3), 2.12 (m, 1H, H-21′), 2.25 (m, 1H, H-22′), 3.57 (m, 2H, H-51′, -52′), 3.75 (m, 1H, H-4′), 4.33 (m, 1H, H-3′), 6.12 (t, 1H, H-1′), 7.29 (s, 1H, H-6), 7.38 (bs, 7H, 3-NH, 5′-OH, exchangeable guanidine). ^{13}C NMR (100 MHz DMSO-d_6) δ 13.66 (CH_3), 36.24 (C-2′), 60.87 (C-3′), 62.32 (C-5′), 83.47 (C-1′), 83.70 (C-4′), 109.68 (C-5), 134.52 (C-6), 157.86 (C-2), 158.99 (guanidinium), 173.96 (C-4).

*Step B: Isolation of Crude AZT **4** from AZT Guanidine Salt **20**.* Water (about 137 kg) was combined with 1 equiv of conc HCl and heated to 75 °C. Then water-damp AZT guanidine **20** cake (about 263 kg; dry weight is 200 kg) was added; first a solution forms, and soon after a precipitate began to form. The mixture was cooled to 5 °C, maintained for 6 h, filtered, and washed with water (3 × 20 kg) to yield 200.5 kg of water-damp crude AZT **4** (dry weight of 136.5 kg, 510.8 mol, 83.3%). HPLC indicated 98.9% AZT **4** and 0.52% dimer **7**.

*Step C: Preparation of Pure AZT **4**.* Water-damp crude AZT 4 (176.2 kg, dry weight 120 kg,) from Step B was dissolved in water (304 kg) and acetone (190 kg) at 50 °C. Active carbon (3 kg) was added and cooled to 30 °C; Celite (9 kg) was added; the mixture was filtered over a Celite bed, and the bed was washed with additional 40 °C acetone (3 × 25 kg). About 74 kg of acetone was distilled at atmospheric pressure and, until ^{1}H NMR determination, produced a water/acetone ratio of 2.55 (acceptable range is 2.0−3.0). The mixture was cooled to 35 °C, seeded with pure AZT **3**, cooled to 23 °C, and agitated for 12 h. The mixture was vacuum-distilled (400−550 mmHg) at 38 °C until 81 kg (little observable distillate) had been collected. The mixture was cooled to 5 °C, maintained for 1 h, a small sample of crystals was filtered, and HPLC determined the dimer content to be 0.16% (end point <0.2%). The mixture was filtered, washed with water (2 × 30 kg) to 137 kg of water damp crystals and vacuum-dried at 55−65 °C to 106.2 kg (88.5%) of AZT **4**; 99.7% HPLC A%, **7** content = 0.15%.

■ ASSOCIATED CONTENT

Supporting Information. Experimental and calculated ^{13}C NMR spectra for neutral and ionized forms of trityl AZT, AZT, and guanidine. This material is available free of charge via the Internet at http://pubs.acs.org.

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