

## Ynamines Derived from Nucleic Acids Bases: Synthesis, Reactivity and Biological Activity

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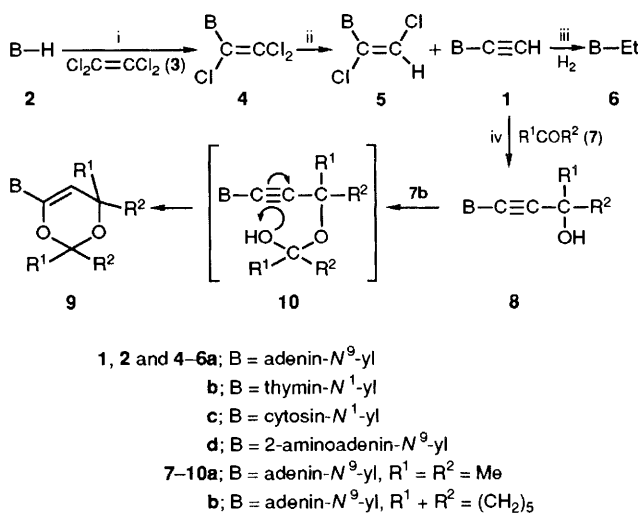
Ynamines **1a–d** are prepared by alkylation of nucleic acids bases with tetrachloroethylene followed by elimination of halogens from the intermediates **4a–d**; reaction of **1a** with acetone and cyclohexanone gives carbinols **8a** and **b** and in the case of **8b** cyclic ketal **9a** is also obtained; compound **1a** is a substrate for adenosine deaminase.

Ynamines have received only scant attention probably because of their limited stability.<sup>1,2</sup> Thus, ynamines derived from aromatic heterocycles and comprising a terminal acetylene moiety have not been reported to the best of our knowledge. Compounds **1** having a nucleic acid base or related moiety attached directly to a reactive but relatively small ethynyl residue are of distinct biological interest as simple nucleoside analogues. In addition, a weaker basicity of such ynamines should enhance their stability, especially toward hydrolysis. Also, compounds **1** could serve possibly as starting materials for unsaturated acyclic nucleoside analogues (see, e.g. refs. 3 and 4).

We now report a general synthesis as well as some chemical and biological properties of ynamines **1a–d**. The sodium salts

generated *in situ* from heterocycles **2a–d** by using NaH (2 equiv.) in hexamethylphosphoric triamide (HMPA) at 60 °C were alkylated with tetrachloroethylene (**3**, 3–6 equiv.) for 15 h (Scheme 1). Trichloroenamines **4a–d** were isolated by flash chromatography on silica gel in 25–30% yields.† The alkylation is highly regioselective giving predominantly *N*<sup>9</sup>-alkylated purines **4a, d** and *N*<sup>1</sup>-substituted pyrimidines **4b, c**. In the instance of adenine derivatives **4a** and **d** only ca. 5% of *N*<sup>7</sup>-isomers were present. The solvent (HMPA) is impor-

† All new compounds were characterized by elemental analyses, IR spectra and, with the exception of **1c**, by UV, NMR and mass spectroscopy.



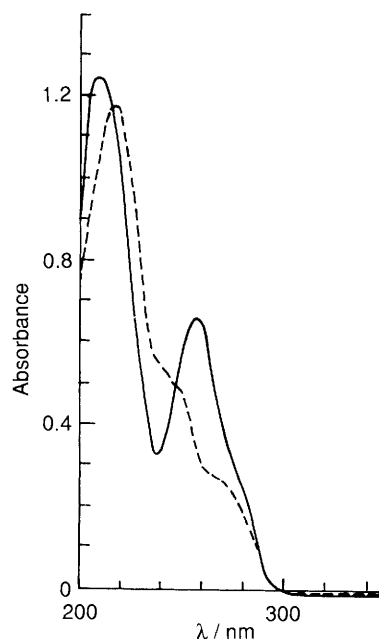
**Scheme 1** Reagents and conditions: i, NaH, HMPA, 60°C; ii, LiBu, THF, -70°C; iii, Pd/C, EtOH; iv, NaNH<sub>2</sub>, THF

tant for successful alkylation. Thus, alkylation of thymine **2b** with tetrachloroethylene **3** in dimethyl sulfoxide (DMSO) led only to a reduction product of **4b**, the dichlorovinyl derivative **5b** in 20% yield.

Compounds **4a-d** were converted to ynamines **1a-d** using 1-butyllithium (LiBu, 4 equiv.) in tetrahydrofuran (THF) at -70°C for 1-3 h in 50% yields. The reaction is accompanied by a regioselective reduction of one chlorine atom of the starting materials **4a, c, d** but not **4b**. Thus, compound **5a** was obtained in 10% yield along with ynamine **1a** and it was identical with a product of alkylation of adenine **2a** with trichloroethylene. Catalytic hydrogenation of **1a** and **c** gave *N*<sup>9</sup>-ethyladenine **6a** and *N*<sup>1</sup>-ethylthymine **6b** with properties identical to those of authentic samples.<sup>5,6</sup> Unlike ynamines derived from strong tertiary bases<sup>1</sup> compounds **1a, b** and **d** are stable in aqueous solutions. Ynamine **1c** is of limited stability although it gave a correct elemental analysis and IR spectrum (KBr). The <sup>1</sup>H and <sup>13</sup>C NMR spectra in (CD<sub>3</sub>)<sub>2</sub>SO at an ambient temperature and (CD<sub>3</sub>)<sub>2</sub>NCDO at -20°C showed, in addition to signals of **1c**, several peaks in the alkenic region indicating the presence of polyacetylene polymer(s).<sup>7</sup> The limited stability of **1c** cannot be attributed solely to the basicity of the cytosine moiety because ynamine **1d** containing a more basic 2-aminoadenine residue is quite stable.

The reaction of ynamine **1a** with acetone **7a** in the presence of NaNH<sub>2</sub> in THF gave the carbinol **8a** in 45% yield. The higher yield (70%) was obtained with cyclohexanone **7b** but compound **8b** was accompanied by cyclic ketal **9a** (30%). The latter became virtually the sole product when more than 2 equiv. of **7b** were employed. Obviously, carbinol **8b** is an intermediate in the process which includes formation of hemiketal **10a** and subsequent cyclization to give **9a**. It is noteworthy that a base-catalysed reaction of diacetylene carbinols with formaldehyde was reported<sup>8</sup> to give dioxolane derivatives also *via* the corresponding hemiacetals.

Ynamine **1a** was deaminated with adenosine deaminase (ADA) under standard conditions.<sup>9</sup> After 3 days *ca.* 90% deamination was observed at room temperature as shown by TLC in CH<sub>2</sub>Cl<sub>2</sub>-MeOH (9:1) and UV spectra (Fig. 1). The *N*<sup>9</sup>-(1-propyn-3-yl)- and *N*<sup>9</sup>-vinyladenine as well as compound **1d** were inert towards ADA. Compound **1a** is the simplest substituted adenine amenable to deamination catalysed with ADA. Compounds **4b, 1a, d** and **8b** inhibited the growth of murine leukaemia L 1210 cells with IC<sub>50</sub> 40, 100, 125 and 150 μmol dm<sup>-3</sup>, respectively, as determined by a clonogenic assay.<sup>10</sup> Compounds **1a, d** and **4a** suppressed the tumour growth in cultures of mouse colon tumour C38 and human lung tumour H8 or H116 at 0.04, 0.5 and 1 mg per disk,



**Fig. 1** Deamination of ynamine **1a** with ADA. The incubation mixture contained 4.7 μmol **1a** ml<sup>-1</sup> and 0.66 enzyme unit ml<sup>-1</sup> in 0.05 mol dm<sup>-3</sup> Na<sub>2</sub>HPO<sub>4</sub>, pH 7.5. An aliquot was diluted with buffer and the UV spectrum was recorded. (—) UV spectrum before addition of ADA, (---) after 3 days of incubation of **1a** with ADA at room temperature.

respectively, as shown by a zone assay.<sup>10</sup> Further investigation of the synthetic utility of ynamines **1a-d** as well as other biological tests are in progress.

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