Synthesis by conjugate radical addition of new heterocyclic amino acids with nucleic acid bases in their side chains

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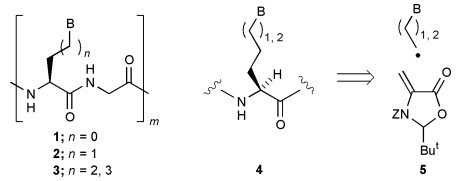
Received (in Cambridge, UK) 21st August 2000, Accepted 20th September 2000
First published as an Advance Article on the web 13th October 2000

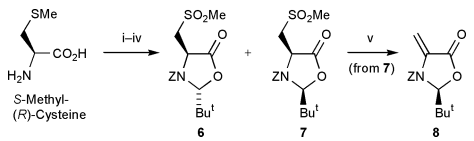
_N_-(2-Iodoethyl) and _N_-(3-iodopropyl)pyrimidines and purines undergo stereoselective conjugate radical addition with an optically active oxazolidinone acceptor to give _syn_-adducts that can be converted into pyrimidine and purine amino acids.

Peptide-based nucleic acid analogues (PNAs) have attracted much attention as molecules with the potential to interact with nucleic acid chains.[1] Suggested applications include antisense properties.[2] Nielsen's PNA has been shown to form duplexes with the complementary DNAs.[1] DNA recognition using analogues with a 'real' peptide backbone has, however, proved more elusive. Substituted alanine oligomers **1** (B = pyrimidine or purine base) and homologues **2** (termed α-PNA[3]) do not demonstrate hybridisation with DNA and insufficient flexibility of the polypeptide chain has been suggested as the cause,[4] whereas triplex formation between tetrapeptides of type **2** and poly(dT) or poly(dU) has been reported.[5] Our interest in unusual amino acids led us to propose the homologous amino acids **4** carrying the nucleic acid bases with a 3- or 4-methylene tether to the peptide backbone, as components for PNA variant **3**. Residues **4** are also analogues of natural pyrimidine and purine amino acids.[6] We report here our flexible methodology based on stereospecific radical chemistry.[7]

In contrast to published routes to residues with C_2 tethers,[3,5,8] we determined to link preformed heterocycles with the peptide backbone by forming a *carbon–carbon* bond in the tether, and proposed to generate the C(β)–C(γ) bond by conjugate radical addition to chiral acceptor **7** (Scheme 1).[9] The (*S*)-acceptor **8** was prepared from *S*-methyl-(*R*)-cysteine (Scheme 2) by adaptation of a published sequence to the *N*-benzoyl analogue.[10] *syn*-Sulfone **7** was formed as a 10:1 mixture with its *anti*-diastereoisomer **6** [57% overall from *S*-methyl-(*R*)-cysteine] and easily separated by column chromatography.[10] The *syn*-configuration was supported *inter alia* by mutual NOE enhancements between C-2(H) and C-4(H). Base treatment afforded (*S*)-oxazolidinone **8** as a crystalline solid.



Scheme 1



Scheme 2 *Reagents*: i, NaOH aq.; Bu^{t}CHO, Dean-Stark; iii, $PhCH_2OCOCl$ (ZCl); iv, oxone®, MeCN–H_2O; v, DBU.

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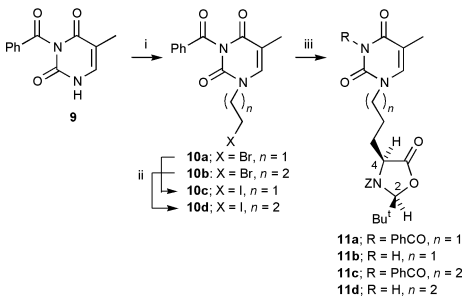
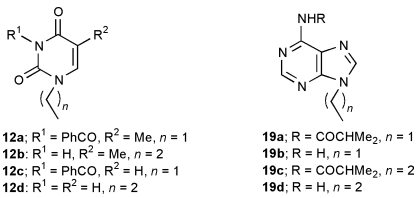
The radical precursors were haloalkyl pyrimidines and purines, prepared from the appropriately protected heterocyclic base and an ω-haloalcohol.[11] Thus 3-benzoylthymine **9**[12] was coupled with 2-bromoethanol or 3-bromopropanol (DIAD, Ph_3P) to afford the 1-(ω-bromoalkyl) derivatives **10a,b**, respectively (Scheme 3). Attempts to generate radicals from these bromides proved fruitless, so they were converted directly to the iodoalkyl compounds **10c,d**, respectively (NaI, propanone reflux; 85, 87% from **9**).

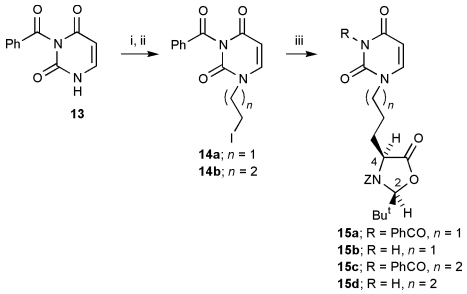


Scheme 3 *Reagents*: i, $HO(CH_2)_{n+1}Br$, $Pr^iO_2CN{=}NCO_2Pr^i$, Ph_3P; ii, NaI, Me_2CO reflux; iii, Method A: **10** (1 mol equiv.), Bu_3SnH (1 mol equiv.), AIBN (0.1 mol equiv.), toluene reflux; Method b: **8** (2 mol equiv.), Bu_3SnCl (0.3 mol equiv.), $NaBH_3CN$ (2 mol equiv.), AIBN (0.1 mol equiv.).

Iodide **10c** was treated under two protocols differing in the method for radical generation;[9] method A: with oxazolidinone **8** (1 mol equiv.) in toluene at reflux containing AIBN (0.1 mol equiv.) and dropwise addition of Bu_3SnH (1 mol equiv.); or method B: with oxazolidinone **8** (2 mol equiv.), Bu_3SnCl (0.3 mol equiv.), $NaBH_3CN$ (2 mol equiv.) and AIBN (0.1 mol equiv.) in *tert*-BuOH at reflux. Method A afforded the conjugate addition product **11a** (26%) and reduction product **12a** (24%), whereas method B after 16 h afforded 54% of conjugate addition products consisting of the adduct **11a** (24%) and the 3-debenzoylated derivative **11b** (30%), with no reduced material. The extent of debenzoylation was time dependent; a reaction time of 40 h led to **11b** as the sole addition product (47%). This suggests the deacylation may be *via* hydride-mediated reduction of the out-of-plane benzoyl carbonyl group, a possibility supported by an observed decrease in debenzoylation when less $NaBH_3CN$ is used in method B, and that debenzoylation of **10** occurs in the presence of $NaBH_3CN$ alone.[13] When 1-iodopropylthymine derivative **10d** was treated under method B, adduct **11c** was not found and deacylated adduct **11d** was isolated (25%) along with reduction product **12b** (75%).

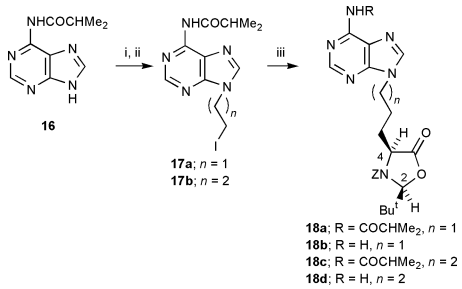
We elected to extend these standard protocols (method B preferred) to other pyrimidines and purines rather than optimise each conjugate addition. Thus 3-benzoyluracil **13**[12] was



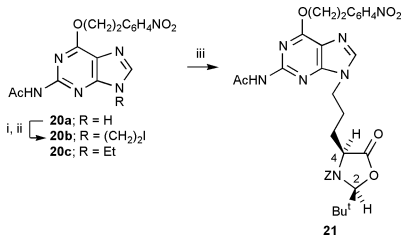


Scheme 4 *Reagents*: i, ii as Scheme 3; iii, Method B.

converted into the 1-(iodoalkyl) derivatives **14a,b** (Scheme 4). Method B applied to **14a** afforded addition product **15a** (41%) and reduction product **12c** (46%); when the reaction was left for 2 days, deacylated addition product **15b** (51%) was isolated. Homologue **14b** gave debenzoylated adduct **15d** (44%) with reduced material **12d** (51%).‡ In the purine series, the 9-(iodoalkyl)adenines **17a,b** were prepared from (2-methylpropionyl)adenine **16**[14] (Scheme 5). Using method B, iodoethyl compound **17a** led to the expected mixture of conjugate addition [40%; acylated **18a** (26%) and deacylated **18b** (14%)] and reduction [36%; acylated **19a** (17%) and deacylated **19b** (19%)]. Iodopropyl derivative **17b** likewise gave adducts [22%; acylated **18c** (12%) and deacylated **18d (**10%)] and reduced compounds [34%; acylated **19c** (11%) and deacylated **19d** (23%)]. Finally, a protected guanine **20a**[15] was converted into the 9-iodoethyl derivative **20b** (Scheme 6) and method A led to adduct **21** (21%) and reduction to **20c** (20%).

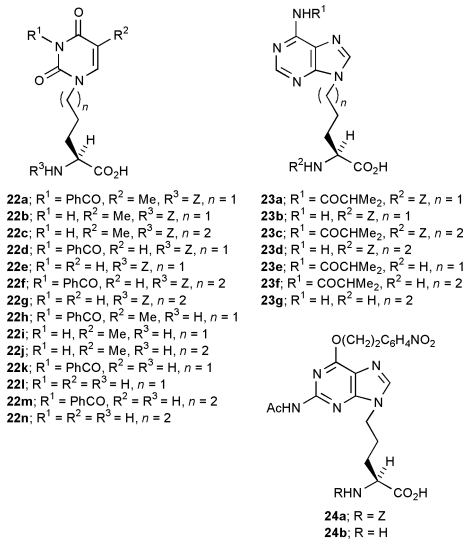


Scheme 5 *Reagents*: i, ii as Scheme 3; iii, Method B.



Scheme 6 *Reagents*: i, ii as Scheme 3; iii, Method A.

The illustrated conjugate radical addition products were all *syn*-adducts, as determined by NOE studies [enhancements between C-2(H) and C-4(H)]. Only one diastereoisomer was visible in the ^{1}H NMR spectra at 300 MHz. All of these *syn*-oxazolidinones could be easily and efficiently converted into *N*-benzyloxycarbonyl-(*S*)-amino acids (suitable for peptide coupling) by base hydrolysis (LiOH, aq. THF, 0 °C, 30–60 min; 70–98%). Thus the three thymine-substituted Z-amino acids **22a–c** (having 3- or 4-carbon tethers for the pyrimidine) were prepared from the adducts **11a**,**b**,**d**, respectively. The uracil Z-amino acids **22d–g** were likewise prepared from adducts **15a–d**, respectively, as were adenine derivatives **23a–d** (from **18a–d**, respectively) and guanine Z-amino acid **24a** (from **21**). To monitor optical purity, the Z group was removed by hydrogenolysis (Pd–C, EtOH–H_2O; 60–80%) to afford the amino



acids **22h–n**, **23e–g** and **24b**, analysed by esterification (AcCl, EtOH, reflux) and subsequent conversion to the Mosher amides (*R*-3,3,3-trifluoro-2-methoxy-2-phenylpropanoyl chloride, pyridine);[16] ^{19}F NMR spectroscopy revealed, *e.g.* 86–91% e.e. for the amino acids **22i**,**j**,**l**,**m**, and **23f**.

We have thus made available a range of novel pyrimidinyl and purinyl amino acids for application, for example, in PNA variants.

We thank the Open University for financial support (competitive studentship to D. J. C. B.) and the EPSRC National Mass Spectrometry Service Centre (Swansea) for some MS data.

Notes and references

‡ The yield of **15d** could be increased to 62% by using 5 mol equiv. of acceptor **8** in method B, but we more usually used 2 mol equiv. of this valuable optically active intermediate. When less than 2 mol equiv. $NaBH_3CN$ was used, some of the benzoylated adduct **15c** was isolated.

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