Nucleotide Base Recognition: Ditopic Binding of Guanine to a Macrocyclic Receptor containing Naphthyridine and Naphthalene Units†

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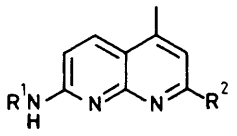
A novel series of receptors for guanine has been prepared and is shown, by ^{1}H n.m.r., to bind the nucleotide base *via* both hydrogen bonding and aromatic stacking interactions.

Recent discoveries of the role played by guanine nucleotide binding proteins (G-proteins) in the function of cell-surface receptors[1] have focused much attention[2] on the biochemical recognition and complexation of guanine. One possible strategy involves coupling hydrogen bonding to the periphery of the purine base with a stacking interaction perpendicular to its plane.[3] This is shown by ribonuclease T_1 which binds guanine selectively *via* two hydrogen bonds from guanine to the peptide backbone and an aromatic stacking interaction between a tyrosine residue and the purine plane.[4] We have recently incorporated this two-point binding strategy into a synthetic receptor showing hydrogen bond complementarity for thymine derivatives.[5] In order to investigate further the molecular recognition of purines and pyrimidines, as well as to develop molecules that bind to specific nucleotide sequences, we sought to construct hosts for other key nucleotide bases.[6] Here we report the synthesis and binding properties of a receptor for guanine that is modelled on the active site of ribonuclease T_1 and contains both hydrogen bonding and hydrophobic recognition sites.

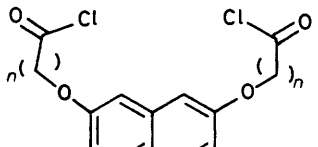
Guanine receptors (**1**) and (**2**) make use of the triply hydrogen-bonded complementarity that exists between 7-amino-1,8-naphthyridines and guanine (**3**). The naphthyridine replaces cytosine in the Watson–Crick base pair by providing the same hydrogen bonding groups (two acceptors, one donor) but in a readily synthesized and functionalized molecule.[7] 2,7-Dialkoxynaphthalenes were chosen as the hydrophobic units as they occur in several DNA intercalating drugs[8] and can be easily incorporated into macrocyclic receptors.[5]

2-Chloro-4-methyl-7-acetylamino-1,8-naphthyridine (**4**) was prepared by condensation (H_3PO_4) of 2,6-diaminopyridine and ethyl acetoacetate, followed by acetylation (Ac_2O) and chlorination ($POCl_3$).[9] Treatment of (**4**) with an excess of sodium 2-hydroxyethoxide gave the amino alcohol (**5**) which was then coupled under high dilution conditions with acid chlorides (**6**)[5] and (**7**),§ to provide naphthyridine macrocycles (**1**) and (**2**)* in 20 and 12% yield, respectively.

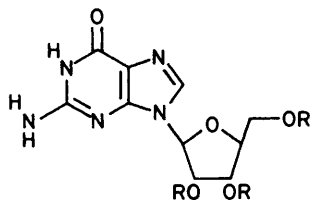
Addition of 2′,3′,5′-tri-*O*-pentanoylguanosine (**8**) (1 equiv.)¶ to a $CDCl_3$ solution of (**1**) caused distinctive changes in the ^{1}H n.m.r. spectrum of both host and guest. The NH resonance of (**1**) and the NH and NH_2 resonances of (**8**) are



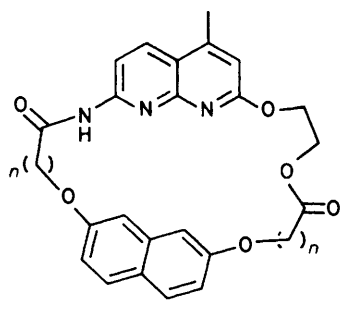
(**4**) R^1 = MeCO, R^2 = Cl
(**5**) R^1 = H, R^2 = OCH_2CH_2OH
(**6**) R^1 = PrCO, R^2 = OEt



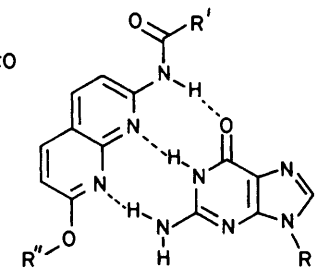
(**6**) n = 3
(**7**) n = 2



(**8**) R = pentanoyl



(**1**) n = 3
(**2**) n = 2



(**3**)

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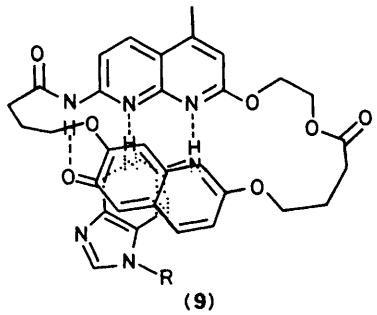
§ Prepared by refluxing 2,7-dihydroxynaphthalene with acrylonitrile followed by methanolysis (MeOH–HCl), hydrolysis (HCl, acetone), and chlorination [$(COCl)_2$].

* All new compounds gave satisfactory spectroscopic, microanalytical, and/or mass spectral analyses.

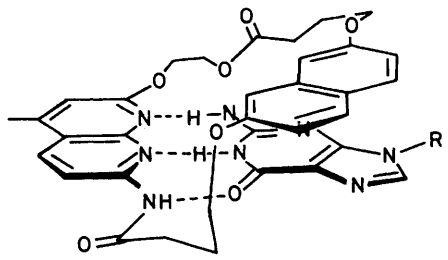
Spectral data for (**1**): ^{1}H n.m.r. ($CDCl_3$) δ 8.82 (1H, br. s, NH), 8.31, 8.18 (2H, 2d, *J* 9 Hz, naphthyr 5-, 6-H), 7.50 (2H, m, naphthal 4-, 5-H), 7.09, 6.90 (2H, 2s, naphthal 1-, 8-H), 6.91 (2H, m, naphthal 3-, 6-H), 6.76 (1H, s, naphthyr 3-H), 4.63 (2H, t, *J* 5 Hz, pyr OCH_2), 4.52 (2H, t, *J* 5 Hz, CH_2OCO), 4.19 (2H, t, *J* 6 Hz, naphthal OCH_2), 3.88 (2H, t, *J* 6 Hz, naphthal OCH_2), 2.60 (3H, s, Me), 2.57 (4H, m, $COCH_2$), 2.35 (2H, m, $COCH_2CH_2$), 2.12 (2H, m, $COCH_2CH$); m.s. *m/z* 515.2041.

For (**2**): ^{1}H n.m.r. ($CDCl_3$) δ 8.99 (1H, br. s, NH), 8.20, 8.12 (2H, 2d, *J* 9 Hz, naphthyr 5-, 6-H), 7.63, 7.54 (2H, 2d, *J* 9 Hz, naphthal 4-, 5-H), 7.54, 6.96 (2H, 2d, *J* 2 Hz, naphthal 1-, 8-H), 7.11, 6.88 (2H, 2dd, *J* 9 and 2 Hz, naphthal 3-, 6-H), 6.73 (1H, s, naphthyr 3-H), 4.92 (2H, t, *J* 8 Hz, pyr OCH_2), 4.60 (4H, m, CH_2OCO, naphthal OCH_2), 4.16 (2H, t, *J* 6 Hz, naphthal OCH_2), 2.80 (4H, m, $COCH_2$), 2.58 (3H, s, Me); m.s. *m/z* 487.1744.

¶ Prepared by reacting guanosine with pentanoyl chloride.[10]



(9)



(10)

R = 2',3',5'-tri-O-pentanoylribose

shifted downfield by 1.36, 0.31, and 0.25 p.p.m., respectively, reflecting the formation of a triply hydrogen-bonded complex [as in (**3**)]. In addition, all six of the naphthalene proton resonances are shifted upfield owing to the close approach of the naphthalene to the bound guanine. Interestingly, the naphthalene-5, -6, and -8 protons experience greater upfield shifts (0.15, 0.14, and 0.18 p.p.m.) than those at positions 4, 3, and 1 (0.1, 0.09, and 0.1 p.p.m.). These results suggest a structure for the complex (**1:8**) [shown in (**9**) and (**10**)] in which, owing to the unsymmetrical nature of (**1**), the naphthalene stacks skew to the guanine, thus placing the protons at positions 5, 6, and 8 closer to the purine and naphthyridine ring currents. The guanine 2-H shifts upfield by 0.06 p.p.m., which is consistent with its distance from the naphthalene ring. Similar shifts are seen in the 1 : 1 complex between (**2**) and (**8**).

Further support for the ditopic recognition of (**8**) by (**1**) and (**2**) comes from their association constants which were determined from ^{1}H n.m.r. titration data using a Foster–Fife analysis.[11] Both (**1**) (K_s 531 dm^3 mol^{-1}) and (**2**) (K_s 712 dm^3 mol^{-1}) show a more than four fold increase in association constant compared to simple naphthyridine (**11**)†† (K_s 126 dm^3 mol^{-1}) which lacks the stacking component. This corresponds to a contribution of ~1 kcal mol^{-1} (1 kcal = 4.184 kJ) from the naphthalene–guanine stacking interaction to the overall binding free energy (~4 kcal mol^{-1}). Similar binding enhancements have been seen in related ditopic receptors for thymine[5] and adenine.[12]

In summary, we have prepared a novel series of receptors which bind guanine derivatives by both hydrogen bonding and hydrophobic stacking interactions. These hosts, in conjunction with receptors for other nucleotide bases,[5] will form the basis of a systematic approach to 'artificial repressors' which may recognize and bind to specific nucleic acid sequences.

Note added in proof: Feibush *et al.*[13] have reported the use of simple amino-1,8-naphthyridines to bind to guanine derivatives.

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†† Prepared by treating (**4**) with sodium ethoxide, followed by reaction with butyryl chloride.