Available online at www.sciencedirect.com
SCIENCE DIRECT
Tetrahedron
Tetrahedron 62 (2006) 3439–3445

Parallel synthesis of an indole-based library via an iterative Mannich reaction sequence

Charlotta Lindquist,[a,b] Oguz Ersoy[b,*] and Peter Somfai[a,*]

[a]*Organic Chemistry, Department of Chemistry, Royal Institute of Technology, S-100 44 Stockholm, Sweden*
[b]*Amersham Biosciences, S-751 84 Uppsala, Sweden*

Received 12 January 2005; revised 12 December 2005; accepted 12 January 2006

Available online 8 February 2006

Abstract—A library of 1,3-disubstituted indoles has been prepared via an iterative Mannich reaction sequence. The first Mannich reaction with secondary amines and formaldehyde preferentially yields 3-aminomethyl indoles, while the second Mannich reaction introduces an additional aminomethyl group at the N1-position of the indole ring. A library of 25 substituted indoles has thus been prepared in moderate to good yields with purity.

1. Introduction

Affinity chromatography is a powerful technique where a ligand with specific affinity for a biological substance, usually a protein, is used for the purification of this substance.[1] Combinatorial synthesis[2] presents itself as a useful tool for the development of new affinity ligands. One such recent campaign has focused on the use of 1,3-substituted indoles as affinity chromatography ligands.

Indoles and related heterocyclic structures are found in numerous natural products with interesting biological activities, and several combinatorial synthesis studies have used the indole heterocycle as a core-function.[3] The N1- and C3-positions of the indole nucleus are relatively electron-rich and react with various types of electrophiles.[4] In the present study, the aim was to introduce functionality in the N1- and C3-positions while leaving the C2-position unsubstituted. For the N1-position, alkylation with an alkyl halide is a plausible option, but controlling the regioselectivity in such reaction can be difficult.[5] For derivatization at the C3-position, a Mannich-type addition was chosen as the lone pair of the resultant amine moiety was expected to participate in the binding interactions of the projected affinity ligands. Due to the known difficulty of selective alkylation at the N1-position, the Mannich reaction at the C3-position with a secondary amine and formaldehyde was chosen as the first step. Although this reaction proceeded efficiently, we were surprised to observe a bis-Mannich product when extended reaction times and excess reagents were applied. More surprisingly, this bis-Mannich product was quite stable under conditions typical to affinity chromatography. A review of the literature revealed only two accounts where similar bis-Mannich products had been observed, in both occasions as byproducts.[6] However, there were no accounts of the use on this dual reactivity to build compounds employing two different Mannich reactions.

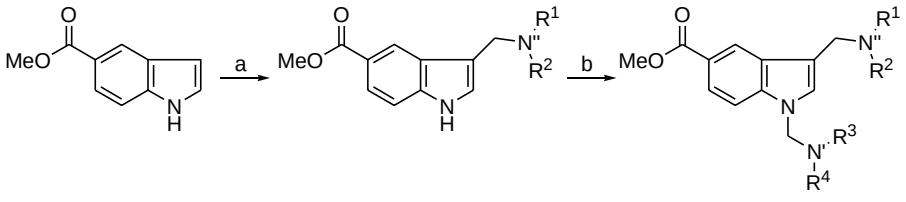
Intrigued by this finding, we undertook the synthesis of an 1,3-aminomethyl indole library where two different Mannich reactions with secondary amines and formaldehyde were used in sequence to produce differentially substituted indole scaffolds. Methyl indole-5-carboxylate (**1**) was selected as the starting material, with the aim of using the carboxylic acid functionality as an attachment point to the chromatography matrix. Herein we describe the synthesis of 25 1,3-aminomethyl indole compounds.

2. Results and discussion

Mannich reactions between indole and secondary imines in water at low temperature are known to substitute the N1 position of indoles.[7] The products, *N*-aminal indoles, are relatively stable but convert to the thermodynamically more stable C3-substituted aminomethyl indoles upon heating at neutral pH or acid treatment at room temperature.[4] Accordingly, we devised a synthetic strategy where the C3-position of **1** was first substituted under standard

Keywords: Library synthesis; Mannich reaction; Affinity ligands.
* Corresponding authors. Tel.: +46 18 6121162; fax: +46 18 6121837 (O.E.); tel.: 46 8 7906960; fax: 46 8 7912333 (P.S.); e-mail addresses: ersoy@amersham.com; somfai@kth.se

0040–4020/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tet.2006.01.047



2 $R^1R^2N''H$=pyrrolidine, 77%
3 $R^1R^2N''H$=4-methylpiperidine, 93%
4 $R^1R^2N''H$=diethylamine, 78%
5 $R^1R^2N''H$=4-methylpiperazine, 45%
6 $R^1R^2N''H$=morpholine, 57%
7 $R^1R^2N''H$=benzylmethylamine, 66%

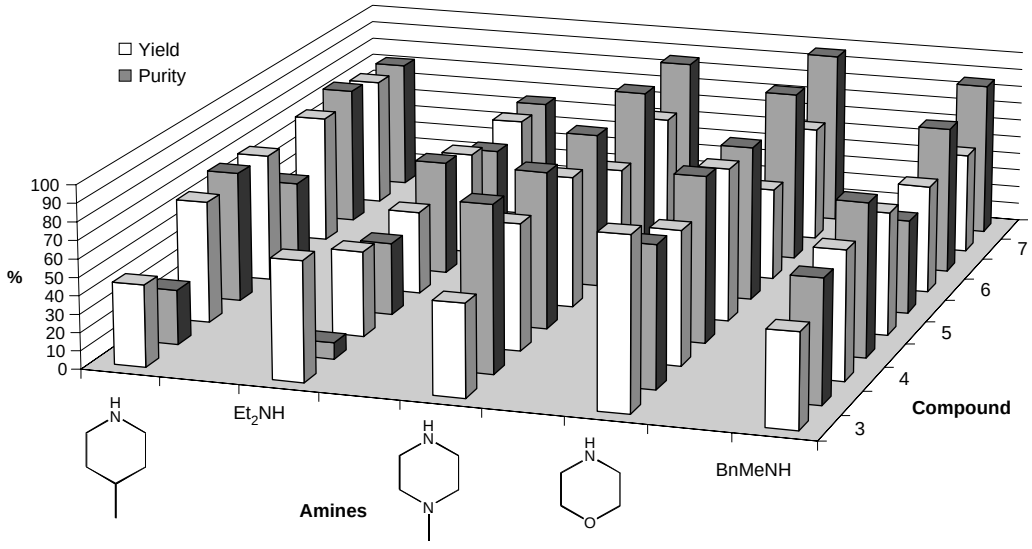
Scheme 1. (a) $R^1R^2N''H$ (1.2 equiv), HCHO (1.2 equiv), room temperature, 18 h; (b) $R^3R^4N'H$ (1.5 equiv), HCHO (1.5 equiv), room temperature, 48 h.

Mannich conditions to yield the corresponding thermodynamically stable products, 3-aminomethyl indoles **2–7** (Scheme 1).

Compounds **3–7** were then selected for further library synthesis. The combination of these compounds with five different secondary amines then gave a library of 25 novel compounds, **8–32** (Tables 1 and 2). The second Mannich reaction required prolonged reaction times to reach completion and all attempts with elevated reaction temperatures resulted in complicated reaction mixtures, perhaps due to scrambling of the secondary amine moieties on the indole scaffold. To simplify the purification of the library members excess amine was removed using a polymer bound electrophilic scavanger, methylisocyanate polystyrene.[11] As can be seen from the data in Table 1, the yields vary from good to moderate, the reason probably being that some product was lost during purification, and that the purities of the library compounds were generally high. However, when Et_2NH was used in the second Mannich reaction, that is, formation of compounds **9**, **14**, **19**, **24** and **29**, products with lower purities were obtained, which might be due to the more sterically hindered amine moiety.

In order to exclude the possibility of scrambling of the N1- and C3-substituents during the second Mannich reaction,

Table 1. Yields and purity of library compounds[a]

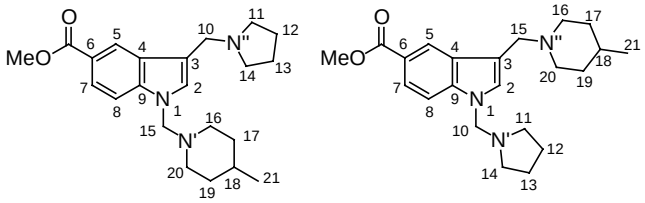


[a] Purity was determined from relative peaks areas of HPLC chromatogram (runs with gradient of 0–100% acetonitrile in water (0.05% TFA) for 10 min at λ= 214 nm). Yield was calculated from weight after removal of excess amine by scavanger resin. Reaction conditions: substrate (1 equiv), R^3R^4NH (1.5 equiv), HCHO (1.5 equiv), dioxane:HOAc, room temperature, 48 h.

Table 2. Compound numbering

Compound	4-Methylpiperidine	Diethylamine	1-Methylpiperazine	Morpholine	Benzylmethylamine
3	**8**	**9**	**10**	**11**	**12**
4	**13**	**14**	**15**	**16**	**17**
5	**18**	**19**	**20**	**21**	**22**
6	**23**	**24**	**25**	**26**	**27**
7	**28**	**29**	**30**	**31**	**32**

compounds **33** and **34** were prepared from **2** (64%) and **3** (62%), respectively, by using the same reaction conditions as those employed for the library synthesis. Each reaction yielded a single detectable Mannich product. To confirm the structures of **33** and **34**, 1D ^{1}H and ^{13}C NMR spectra together with 2D $^{1}H,^{1}H$-COSY, $^{1}H,^{13}C$-HMQC[8] and $^{1}H,^{13}C$-HMBC[9,10] spectra were recorded and interpreted. First, assignments of all proton and carbon signals in **33** and **34** were made with assistance of the 2D COSY and HMQC spectra. Secondly, information about long-range ^{1}H, ^{13}C couplings extracted from the HMBC spectra was used to determine the linkage position of the piperidine and pyrrolidine groups on the indole ring. For both compounds the identity of the methylene group C15 was determined by the $^{3}J_{HC}$ coupling between H15 and C16/C20. In a similar way, the identity of the C10 methylene moiety was determined by the $^{3}J_{HC}$ coupling between H10 and C11/C14. In compound **33**, the position of the piperidine group was deduced by the $^{3}J_{HC}$ couplings between H15 and C2/C9 and the position of the pyrrolidine group was deduced by the $^{2}J_{HC}$ coupling between H10 and C3 together with the $^{3}J_{HC}$ couplings between H10 and C2/C4. In compound **34** the situation was reversed, H15 now showed a $^{2}J_{HC}$ coupling to C3 and a $^{3}J_{HC}$ couplings to C2 and C4 while H10 showed $^{3}J_{HC}$ couplings to C2 and C9, thus confirming the postulated structures and verifying that no scrambling of the N1- and C3-substituents occur during the second Mannich reaction (Fig. 1).



33 **34**

Figure 1. Structure and numbering of compounds **33** and **34**.

3. Conclusions

A library of 25 novel bis-1,3-aminomethyl indole compounds has been prepared in moderate to good yields by sequential use of two Mannich reactions. The average purity of the library is greater than 70%. It have also been shown that no scrambling of the amino substituents between the N1- and C3-positions occurs during the second Mannich reaction. Despite the modest library size, these results show that iterative Mannich reactions with different secondary amines and formaldehyde, is a useful method to build libraries of 1,3-aminomethyl indoles. The mild reaction conditions that are employed make this a particularly attractive reaction sequence.

4. Experimental

4.1. Material and methods

Reactions were performed in individual glass tubes placed in IKA-VIBRAX-VXR parallel agitation equipment. Evaporation of solvents was done parallel in a centrifugal evaporator (Speed Vac SC201A, Savant). Scavanger reactions were performed in individual polypropylene tubes (PD-10 columns, Amersham Biosciences). All chemicals and solvents were obtained from commercial sources and were used as received. NMR spectra were obtained on a Bruker Avance 300 in using $CDCl_3$ as solvent and shifts are reported downfield from $(CH_3)_4Si$ (δ 0). LS-MS were obtained on a Hewlett Packard HP1100 MSD (ESI, positive mode) using an YMC C_{18}-hydrosphere column (flow rate: 0.5 mL/min, gradient: 0–100% acetonitrile in water (0.05% TFA) for 10 min, $\lambda=214$ nm). The purity of all compounds were analyzed by analytic RP-HPLC on a Shimadzu 10A system using an ACE C_{18}-column (flow rate: 2 mL/min, gradient: 0–100% acetonitrile in water (0.05% TFA) for 10 min, $\lambda=214$ nm).

4.2. General procedures and spectral data

4.2.1. Typical procedure for the first Mannich reaction. Methyl 3-((pyrrolidin-1-yl)methyl)-1*H*-indole-5-carboxylate (2). To a solution of **1** (300 mg, 1.71 mmol) in dioxane–HOAc (4/1, 2.0 mL) was added a mixture of pyrrolidine (170 μL, 2.06 mmol) and formaldehyde (154 μL, 2.06 mmol, 37 wt% solution in water) in dioxane–HOAc (4/1, 2.0 mL). The resultant mixture was agitated at room temperature overnight. Evaporation of the solvents and flash chromatography (CH_2Cl_2/MeOH/Et_3N, v/v/v 90:10:1) gave **2**·HOAc (419 mg, 77%). HPLC-MS (ESI^+): $t_R=4.34$ min, *m/z* 259 ($M+H^+$, 50%), 188 (100); δ_H (300 MHz, $CDCl_3$) 2.03 (m, 4H, C*H*HCH$_2$N′CH$_2$C*H*H), 2.30 (m, 4H, CH$_2$N′CH$_2$), 3.92 (s, 3H, OMe), 4.52 (s, 2H, CH$_2$N′), 7.49 (d, 1H, $J=8.7$ Hz, H8), 7.61 (s, 1H, H2), 7.88 (dd, 1H, $J=8.7$, 1.5 Hz, H7), 8.52 (d, 1H, $J=1.5$ Hz, H5); δ_C (75 MHz, $CDCl_3$) 23.0, 47.9, 51.9, 52.0, 105.7, 111.8, 120.5, 122.2, 123.4, 126.9, 129.4, 138.8, 168.0; IR (film) 2953, 1709, 1621 cm^{-1}.

4.2.2. Methyl 3-((4-methylpiperidin-1-yl)methyl)-1*H*-indole-5-carboxylate (3). Prepared from **1** (200 mg, 1.14 mmol), 4-methylpiperidine (162 μL, 1.37 mmol) and formaldehyde (103 μL, 1.37 mmol, 37 wt% solution in water) as described for compound **2** to give **3** (304 mg, 93%). HPLC-MS (ESI^+): $t_R=4.81$ min, *m/z* 287 ($M+H^+$, 100%), 188 (58); δ_H (300 MHz, $CDCl_3$) 0.96 (d, 3H, $J=5.7$ Hz, CHC*H*$_3$), 1.51 (m, 3H, C*H*HCH$_2$N′CH$_2$C*H*H), 1.71 (m, 2H, C*H*HCH$_2$N′CH$_2$C*H*H), 2.42 (m, 2H, C*H*HN′C*H*H), 3.27 (m, 2H, CH*H*N′CH*H*), 3.94 (s, 3H, OMe), 4.09 (s, 2H, CH$_2$N′), 7.43 (d, 1H, $J=8.7$ Hz, H8), 7.57 (s, 1H, H2), 7.91 (dd, 1H, $J=8.7$, 1.5 Hz, H7), 8.35 (d, 1H, $J=1.5$ Hz, H5); δ_C (75 MHz, $CDCl_3$) 20.8, 30.8, 33.8, 51.0, 51.9, 52.0, 104.6, 111.8, 120.6, 122.2, 123.4, 127.4, 130.0, 138.8, 168.1; IR (film) 2956, 1697, 1621 cm^{-1}.

4.2.3. Methyl 3-((diethylamino)methyl)-1*H*-indole-5-carboxylate (4). Prepared from **1** (300 mg, 1.71 mmol), diethylamine (212 μL, 2.06 mmol) and formaldehyde (154 μL, 2.06 mmol, 37 wt% solution in water) as described for compound **2** to give **4** (349 mg, 78%). HPLC-MS (ESI^+): $t_R=4.30$ min, *m/z* 261 ($M+H^+$, 23%), 188 (100); δ_H (300 MHz, $CDCl_3$) 1.25 (m, 6H, $J=7.5$ Hz, N′CH$_2$C*H*$_3$), 2.94 (m, 4H, $J=7.5$ Hz, N′C*H*$_2$-CH$_3$), 3.92 (s, 3H, OMe), 4.20 (s, 2H, CC*H*$_2$N′), 7.42 (d,

1H, J=8.7 Hz, H8), 7.58 (s, 1H, H2), 7.88 (dd, 1H, J= 8.7, 1.5 Hz, H7), 8.34 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 8.6, 44.9, 46.4, 52.0, 104, 6, 111.8, 120.3, 122.2, 123.4, 127.2, 129.9, 138.8, 168.0; IR (film) 2952, 1704, 1622 cm^{-1}.

4.2.4. Methyl 3-((4-methylpiperazin-1-yl)methyl)-1*H*-indole-5-carboxylate (5). Prepared from **1** (300 mg, 1.71 mmol), 1-methylpiperazine (228 μL, 2.06 mmol) and formaldehyde (154 μL, 2.06 mmol, 37 wt% solution in water) as described for compound **2** to give **5** (231 mg, 45%). HPLC-MS (ESI^+): t_R=3.74 min, m/z 288 ($M+H^+$, 100%), 188 (87), 130 (8); δ_H (300 MHz, $CDCl_3$) 2.52 (s, 3H, NCH_3), 2.75–3.00 (m, 8H, $CH_2CH_2N'CH_2CH_2$), 3.94 (s, 3H, OMe), 3.98 (s, 2H, CCH_2N'), 7.46 (s, 1H, H2), 7.47 (d, 1H, J=8.7 Hz, H8), 7.91 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.40 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 43.8, 50.0, 51.9, 52.0, 52.7, 109.9, 111.2, 121.8, 121.9, 123.5, 127.0, 127.2, 139.0, 168.2; IR (film) 2954, 1706, 1619 cm^{-1}.

4.2.5. Methyl 3-(morpholinomethyl)-1*H*-indole-5-carboxylate (6). Prepared from **1** (300 mg, 1.71 mmol), morpholine (179 μL, 2.06 mmol) and formaldehyde (154 μL, 2.06 mmol, 37 wt% solution in water) as described for compound **2** to give **6** (324 mg, 57%). HPLC-MS (ESI^+): t_R=4.20 min, m/z 275 ($M+H^+$, 10%), 188 (100); δ_H (300 MHz, $CDCl_3$) 2.73 (m, 4H, $CH_2N'CH_2$), 3.73 (m, 4H, CH_2OCH_2), 3.92 (s, 3H, OMe), 3.96 (s, 2H, CH_2N'), 7.41 (d, 1H, J=8.7 Hz, H8), 7.45 (s, 1H, H2), 7.84 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.47 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 43.3, 51.9, 52.0, 64.4, 108.8, 111.3, 121.5, 122.0, 123.5, 127.5, 127.6, 138.8, 168.1; IR (film) 2948, 1696, 1566, 1112 cm^{-1}.

4.2.6. Methyl 3-((*N*-benzyl-*N*-methylamino)methyl)-1*H*-indole-5-carboxylate (7). Prepared from **1** (50 mg, 285 μmol), *N*-benzylmethylamine (44 μL, 343 μmol) and formaldehyde (26 μL, 343 μmol, 37 wt% solution in water) as described for compound **2** to give **7** (58 mg, 66%). HPLC-MS (ESI^+): t_R=5.47 min, m/z 309 ($M+H^+$, 10%), 188 (27), 122 (100); δ_H (300 MHz, $CDCl_3$) 2.22 (s, 3H, $N'CH_3$), 3.57 (s, 2H, $N'CH_2Ph$), 3.74 (s, 2H, CH_2N'), 3.94 (s, 3H, OMe), 7.12–7.43 (m, 7H, H2, H8, Ar), 7.88 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.50 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 38.5, 50.2, 52.0, 59.1, 106.4, 111.6, 121.0, 122.2, 123.5, 127.3, 129.0, 129.1, 129.2, 130.7, 131.4, 138.9, 168.1; IR (film) 2950, 1698, 1622 cm^{-1}.

4.2.7. Typical procedure for the second Mannich reaction. Compound 23. To a solution of compound **6** (40.0 mg, 145.8 μmol) in dioxane–HOAc (4/1 v/v, 0.5 mL) was added a mixture of 4-methylpiperidine (25.9 μL, 218.7 μmol) and formaldehyde (24.3 μL, 218.7 μmol, 37 wt% solution in water) in dioxane–HOAc (4/1, 0.5 mL). The resultant mixture was agitated for 48 h. The solvents were removed and the residue dissolved in CH_2Cl_2 (1 mL) and added to a slurry of methyl isocyanate polystyrene HL (99 mg, 219 μmol, prewashed and pre-swollen) in CH_2Cl_2 (1 mL). The resultant slurry was agitated for 2 h. Filtration, washing of the resin and removal of the solvents gave compound **23** that was characterized by ^1H NMR and LC-MS. HPLC-MS (ESI^+): t_R=3.91 min, m/z 408 ($M+Na^+$, 61%), 444 (12), 424 (7), 297 (8), 287 (13), 202 (7), 194 (34), 188 (100), 150 (11), 130 (6); δ_H (300 MHz, $CDCl_3$) 0.88 (d, 3H, J=5.7 Hz, $CHCH_3$), 1.22 (m, 3H, $CHHCH(CH_3)CHH$), 1.60 (m, 2H, $CHHCH(CH_3)CHH$), 2.12 (m, 2H, $CHHN''CHH$), 2.55 (m, 4H, $CH_2N'CH_2$), 2.88 (m, 2H, $CHHN''CHH$), 3.75 (m, 4H, CH_2OCH_2), 3.79 (s, 2H, CH_2N'), 3.95 (s, 3H, OMe), 4.82 (s, 2H, CH_2N'), 7.22 (s, 1H, H2), 7.44 (d, 1H, J=8.7 Hz, H8), 7.92 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.46 (d, 1H, J= 1.5 Hz, H5); δ_H (75 MHz, $CDCl_3$) 22.2, 30.7, 34.5, 51.6, 52.3, 53.7, 53.8, 67.0, 68.9, 110.3, 121.9, 122.7, 123.6, 128.4, 130.5, 140.3, 168.5; HRMS (FAB+) calculated for $C_{22}H_{32}N_3O_3$ ($M+H$): 386.2444, found: 386.2428.

4.2.8. Compound 8. Prepared as outlined for compound **23** in 45% yield. HPLC-MS (ESI^+): t_R=4.76 min, m/z 398 ($M+H^+$, 38%), 456 (10), 359 (7), 299 (16), 287 (39), 199 (48), 188 (23), 150 (11), 142 (100), 128 (84); δ_H (300 MHz, $CDCl_3$) 0.87 (d, 3H, J=5.7 Hz, $CHCH_3$), 0.98 (d, 3H, J= 6.3 Hz, $CHCH_3$), 1.60 (m, 6H, $CHHCH(CH_3)CHH$), 1.75 (m, 4H, $HHCCH(CH_3)CHH$), 2.12 (m, 2H, $H_2CCH_2N'CH_2CH_2$), 2.58 (m, 2H, $CHHN'CHH$), 2.87 (m, 2H, $CHHN''CHH$), 3.38 (m, 2H, $CHHN''CHH$), 3.95 (s, 3H, OMe), 4.21 (s, 2H, CH_2N'), 4.86 (s, 2H, CH_2N''), 7.51 (d, 1H, J=8.7 Hz, H8), 7.64 (s, 1H, H2), 7.85 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.32 (d, J=1.5 Hz, 1H, H5); δ_C (75 MHz, DMSO) 21.0, 21.7, 29.8, 30.3, 33.5, 33.7, 45.9, 50.3, 51.7, 67.4, 111.6, 120.6, 121.4, 121.5, 122.3, 127.1, 138.7, 167.3; IR (film) 2974, 1711, 1614 cm^{-1}; HRMS (FAB+) calculated for $C_{24}H_{36}N_3O_2$ ($M+H$): 398.2808, found: 398.2808.

4.2.9. Compound 9. Prepared as outlined for compound **23** in 66% yield. HPLC-MS (ESI^+): t_R=3.99 min, m/z 372 ($M+H^+$, 19%), 430 (15), 404 (38), 348 (24), 299 (23), 287 (100), 202 (16), 188 (46), 150 (32), 122 (10); δ_H (300 MHz, $CDCl_3$) 0.97 (d, 3H, J=5.7 Hz, $CHCH_3$), 1.30 (t, 6H, J= 7.5 Hz, $N''CH_2CH_3$), 1.40–1.83 (m, 5H, $H_2CCH(CH_3)CH_2$), 2.60 (m, 2H, $H_2CCHN'CH_2CH_2$), 3.05 (q, 4H, J=7.5 Hz, NCH_2CH_3), 3.39 (m, 2H, $CHHN''CHH$), 3.93 (s, 3H, OMe), 4.24 (s, 2H, CH_2N'), 4.82 (s, 2H, CH_2N''), 7.47 (d, 1H, J= 8.7 Hz, H8), 7.63 (s, 1H, H2), 7.85 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.28 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 21.3, 23.7, 29.5, 31.5, 51.3, 52.1, 52.4, 107.0, 112.3, 121.0, 122.6, 123.7, 127.9, 130.2, 139.2, 168.5; IR (film) 2956, 1702, 1660 cm^{-1}.

4.2.10. Compound 10. Prepared as outlined for compound **23** in 51% yield. HPLC-MS (ESI^+): t_R=4.27 min, m/z 399 ($M+H^+$, 100%), 457 (14), 287 (17), 212 (13), 200 (37), 188 (83), 142 (8); δ_H (300 MHz, $CDCl_3$) 0.96 (d, 3H, J= 5.7 Hz, $CHCH_3$), 1.52 (m, 3H, $CHHCH(CH_3)CHH$), 1.71 (m, 2H, $CHHCH(CH_3)CHH$), 2.25 (s, 3H, NCH_3), 2.40 (m, 2H, $CH_2N(CH_2C)CH_2$), 2.45 (m, 4H, $CH_2N(CH_3)CH_2$), 2.58 (m, 4H, $CH_2N''CH_2$), 3.28 (m, 2H, $CHHN'CH_3H$), 3.95 (s, 3H, OMe), 4.09 (s, 2H, CH_2N'), 4.85 (s, 2H, CH_2N''), 7.48 (d, 1H, J=8.7 Hz, H8), 7.54 (s, 1H, H2), 7.93 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.32 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 21.6, 30.2, 32.6, 46.1, 50.5, 51.9, 52.4, 52.8, 54.9, 68.5, 110.7, 121.7, 122.6, 123.8, 128.7, 132.6, 139.8, 168.3; IR (film) 2949, 1708, 1620 cm^{-1}; HRMS (FAB+) calculated for $C_{23}H_{35}N_4O_2$ ($M+H$): 399.2760, found: 399.2757.

4.2.11. Compound 11. Prepared as outlined for compound **23** in 95% yield. HPLC-MS (ESI): t_R=4.61 min, *m*/*z* 386 (M+H$^+$, 100%); δ_H (300 MHz, $CDCl_3$) 0.98 (d, 3H, *J*=5.7 Hz, CHC*H*$_3$), 1.25 (m, 2H, C*H*$_2$N CH$_3$), 1.48–1.82 (m, 5H, *H*$_2$CC*H*(CH$_3$)C*H*$_2$), 2.36 (m, 2H, C*H*$_2$N″C*H*$_2$), 2.40 (m, 2H, C*H*$_2$N(CH$_2$C)C*H*$_2$), 2.45 (m, 4H, C*H*$_2$N(CH$_3$)C*H*$_2$), 2.54 (m, 4H, C*H*$_2$N′C*H*$_2$), 2.58 (m, 4H, C*H*$_2$N″C*H*$_2$), 2.69 (m, 4H, C*H*$_2$OC*H*$_2$), 3.95 (s, 3H, OMe), 4.20 (s, 2H, CH$_2$N′), 4.83 (s, 2H, CH$_2$N″), 7.50 (d, 1H, *J*=8.7 Hz, H8), 7.70 (s, 1H, H2), 7.94 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.32 (d, 1H, *J*=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 21.4, 30.1, 31.8, 51.1, 51.3, 52.5, 67.0, 69.0, 106.2, 110.9, 121.3, 123.0, 124.0, 128.7, 133.8, 139.7, 168.2; IR (film) 2953, 1711, 1615, 1115 cm^{-1}; HRMS (FAB+) calculated for $C_{22}H_{32}N_3O_3$ (M+H): 386.2444, found: 386.2448.

4.2.12. Compound 12. Prepared as outlined for compound **23** in 52% yield. HPLC-MS (ESI$^+$): t_R=4.94 min, *m*/*z* 420 (M+H$^+$, 19%), 287 (13), 233 (7), 188 (42), 134 (100); δ_H (300 MHz, $CDCl_3$) 0.93 (d, 3H, *J*=5.7 Hz, CHC*H*$_3$), 1.58 (m, 2H, C*H*HCH(CH$_3$)C*H*H), 1.68 (m, 3H, C*H*HC*H*(CH$_3$)-C*H*H), 2.25 (s, 3H, N″CH$_3$), 2.50 (m, 2H, C*H*HN′C*H*H), 3.33 (m, 2H, C*H*HN′C*H*H), 3.62 (s, 2H, N″C*H*$_2$Ph), 3.95 (s, 3H, OMe), 4.18 (s, 2H, CH$_2$N′), 4.90 (s, 2H, CH$_2$N″), 7.25–7.35 (m, 5H, Ar), 7.40 (d, 1H, *J*=8.7 Hz, H8), 7.64 (s, 1H, H2), 7.93 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.33 (d, 1H, *J*=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 20.9, 29.7, 31.1, 40.2, 51.1, 51.8, 52.0, 59.0, 67.6, 104.9, 110.7, 120.4, 122.4, 123.5, 127.4, 128.4, 128.5, 133.2, 137.9, 139.3, 168.0; IR (film) 2951, 1716, 1617, 1251 cm^{-1}; HRMS (FAB+) calculated for $C_{26}H_{34}N_3O_2$ (M+H): 420.2651, found: 420.2652.

4.2.13. Compound 13. Prepared as outlined for compound **23** in 67% yield. δ_H (300 MHz, $CDCl_3$) 0.87 (d, 3H, *J*=5.7 Hz, CHC*H*$_3$), 1.25 (m, 3H, C*H*HC*H*(CH$_3$)C*H*H), 1.30 (tr, 6H, *J*=7.5 Hz, N′CH$_2$*CH*$_3$), 2.12 (m, 2H, C*H*HCH(CH$_3$)C*H*H), 2.85 (m, 2H, C*H*HN′C*H*H), 3.05 (q, 4H, *J*=7.5 Hz, N′C*H*$_2$CH$_3$), 3.95 (s, 3H, OMe), 4.29 (s, 2H, CH$_2$N′), 4.88 (s, 2H, CH$_2$N″), 7.52 (d, 1H, *J*=8.7 Hz, H8), 7.62 (s, 1H, H2), 7.93 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.33 (d, 1H, *J*=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 10.7, 22.2, 30.7, 34.4, 46.1, 47.1, 51.6, 52.3, 69.0, 109.6, 110.6, 121.8, 122.2, 123.6, 128.4, 131.9, 140.0, 168.4; IR (film) 2949, 1716, 1614 cm^{-1}; HRMS (FAB+) calculated for $C_{22}H_{34}N_3O_2$ (M+H): 372.2651, found: 372.2662.

4.2.14. Compound 14. Prepared as outlined for compound **23** in 47% yield. δ_H (300 MHz, $CDCl_3$) 1.06 (t, 6H, *J*=7.2 Hz, NCH$_2$NCH$_2$*CH*$_3$), 1.30 (tr, 6H, *J*=7.5 Hz, N′CH$_2$-*CH*$_3$), 2.62 (q, 4H, *J*=7.2 Hz, N″C*H*$_2$CH$_3$), 3.00 (q, 4H, *J*=7.5 Hz, N′C*H*$_2$CH$_3$), 3.93 (s, 3H, OMe), 3.94 (s, 2H, CH$_2$N′), 4.30 (s, 2H, CH$_2$N″), 7.39 (d, 1H, *J*=8.7 Hz, H8), 7.65 (s, 1H, H2), 7.89 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.45 (d, 1H, *J*=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 9.1, 12.4, 45.2, 45.3, 46.6, 46.8, 52.2, 70.2, 105.7, 112.1, 120.6, 122.4, 123.6, 127.5, 132.6, 139.0, 168.2; IR (film) 2950, 1709, 1263 cm^{-1}; HRMS (FAB+) calculated for $C_{20}H_{32}N_3O_2$ (M+H): 346.2495, found: 346.2483.

4.2.15. Compound 15. Prepared as outlined for compound **23** in 70% yield. δ_H (300 MHz, $CDCl_3$) 1.30 (tr, 6H, *J*=7.5 Hz, N′CH$_2$*CH*$_3$), 2.36 (s, 3H, NC*H*$_3$), 2.61 (m, 8H, C*H*$_2$C*H*$_2$N″C*H*$_2$C*H*$_2$), 3.02 (q, 4H, *J*=7.5 Hz, N′C*H*$_2$CH$_3$), 3.94 (s, 3H, OMe), 4.29 (s, 2H, CH$_2$N′), 4.82 (s, 2H, CH$_2$N″), 7.48 (d, 1H, *J*=8.7 Hz, H8), 7.65 (s, 1H, H2), 7.94 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.33 (d, 1H, *J*=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 10.0, 45.6, 45.8, 46.7, 50.2, 52.4, 54.7, 68.4, 108.2, 110.7, 121.6, 122.6, 128.5, 123.9, 132.5, 139.7, 168.3; IR (film) 2951, 1704, 1616 cm^{-1}; HRMS (FAB+) calculated for $C_{21}H_{33}N_4O_2$ (M+H): 373.2604, found: 373.2610.

4.2.16. Compound 16. Prepared as outlined for compound **23** in 74% yield. δ_H (300 MHz, $CDCl_3$) 1.28 (t, 6H, *J*=7.2 Hz, N′CH$_2$*CH*$_3$), 2.55 (t, 4H, *J*=4.5 Hz, C*H*$_2$N″C*H*$_2$), 2.95 (q, 4H, *J*=7.2 Hz, N′C*H*$_2$CH$_3$), 3.69 (t, 4H, *J*=4.5 Hz, C*H*$_2$OC*H*$_2$), 3.94 (s, 3H, OMe), 4.22 (s, 2H, CH$_2$N′), 4.82 (s, 2H, CH$_2$N″), 7.48 (d, 1H, *J*=8.7 Hz, H8), 7.59 (s, 1H, H2), 7.94 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.36 (d, 1H, *J*=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 10.5, 46.0, 47.1, 51.1, 52.4, 67.0, 68.8, 109.9, 110.5, 121.9, 122.5, 123.8, 128.6, 131.7, 139.8, 168.3; IR (film) 2967, 1709, 1614, 1116 cm^{-1}; HRMS (FAB+) calculated for $C_{20}H_{30}N_3O_3$ (M+H): 360.2287, found: 360.2289.

4.2.17. Compound 17. Prepared as outlined for compound **23** in 71% yield. δ_H (300 MHz, $CDCl_3$) 1.29 (t, 6H, *J*=7.5 Hz, N′CH$_2$*CH*$_3$), 2.27 (s, 3H, N″C*H*$_3$), 2.95 (q, 4H, *J*=7.5 Hz, N′C*H*$_2$CH$_3$), 3.62 (s, 2H, N″C*H*$_2$Ph), 3.92 (s, 3H, OMe), 4.24 (s, 2H, CH$_2$N′), 4.90 (s, 2H, CH$_2$N″), 7.22–7.38 (m, 5H, Ar), 7.40 (d, 1H, *J*=8.7 Hz, H8), 7.62 (s, 1H, H2), 7.92 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.35 (d, 1H, *J*=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 10.7, 40.6, 46.1, 47.2, 52.4, 59.4, 67.8, 109.8, 110.7, 121.9, 122.3, 123.7, 127.8, 128.6, 128.7, 128.9, 129.1, 131.7, 138.4, 139.9, 168.4; IR (film) 2950, 1709, 1614, 1247 cm^{-1}; HRMS (FAB+) calculated for $C_{24}H_{32}N_3O_2$ (M+H): 394.2495, found: 394.2494.

4.2.18. Compound 18. Prepared as outlined for compound **23** in 71% yield. HPLC-MS (ESI): t_R=3.62 min, *m*/*z* 421 (M+Na$^+$, 13%), 202 (34), 150 (100), 128 (6); δ_H (300 MHz, $CDCl_3$) 0.89 (d, 3H, *J*=5.7 Hz, CHC*H*$_3$), 1.62 (m, 3H, *H*$_2$CC*H*(CH$_3$)C*H*$_2$), 2.19 (m, 2H, *H*$_2$CCH(CH$_3$)-C*H*$_2$), 2.62 (s, 3H, NC*H*$_3$), 2.88–3.18 (m, 10H, C*H*HN″C*H*H, C*H*$_2$C*H*$_2$N′C*H*$_2$C*H*$_2$), 3.94 (s, 2H, CH$_2$N′), 3.95 (s, 3H, OMe), 4.89 (s, 2H, CH$_2$N″), 7.39 (d, 1H, *J*=8.7 Hz, H8), 7.61 (s, 1H, H2), 7.92 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.37 (d, 1H, *J*=1.5 Hz, H5); IR (film) 2951, 1708, 1617 cm^{-1}; HRMS (FAB+) calculated for $C_{23}H_{35}N_4O_2$ (M+H): 399.2760, found: 399.2762.

4.2.19. Compound 19. Prepared as outlined for compound **23** in 46% yield. HPLC-MS (ESI): t_R=3.43 min, *m*/*z* 395 (M+Na$^+$, 31%), 431 (21), 348 (13), 288 (20), 217 (6), 202 (78), 150 (100), 130 (15), 120 (5); δ_H (300 MHz, $CDCl_3$) 1.40 (tr, 6H, *J*=7.2 Hz, N′CH$_2$C*H*$_3$), 2.56 (s, 3H, NCH$_3$), 3.10 (q, 4H, *J*=7.2 Hz, NC*H*$_2$CH$_3$), 2.80–3.20 (m, 8H, C*H*$_2$C*H*$_2$N″C*H*$_2$C*H*$_2$), 3.92 (s, 2H, CH$_2$N′), 3.93 (s, 3H, OMe), 4.06 (s, 2H, CH$_2$N″), 7.28 (s, 1H, H2), 7.52 (d, 1H, *J*=8.7 Hz, H8), 7.88 (dd, 1H, *J*=8.7, 1.5 Hz, H7), 8.35 (d, 1H, *J*=1.5 Hz, H5); IR (film) 2717, 1697, 1621 cm^{-1}.

4.2.20. Compound 20. Prepared as outlined for compound **23** in 73% yield. HPLC-MS (ESI): t_R=3.52 min, *m*/*z* 422 (M+Na$^+$, 31%), 458 (15), 438 (8), 400 (8), 348 (6), 288

(30), 200 (8), 188 (100), 130 (12); δ_H (300 MHz, $CDCl_3$) 1.42 (s, 3H, N″$CH_2CH_2NCH_3$), 2.59 (s, 3H, N′CH_2CH_2-NCH_3), 2.70 (m, 8H, CH_2CH_2N″CH_2CH_2), 2.82–3.02 (m, 8H, CH_2CH_2N′CH_2CH_2), 3.91 (s, 2H, CH_2N′), 3.95 (s, 3H, OMe), 4.82 (s, 2H, CH_2N″), 7.32 (s, 1H, H2), 7.43 (d, 1H, J=8.7 Hz, H8), 7.92 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.49 (d, 1H, J=1.5 Hz, H5); IR (film) 2953, 1701, 1615 cm^{-1}; HRMS (FAB+) calculated for $C_{22}H_{34}N_5O_2$ (M+H): 400.2713, found: 400.2712.

4.2.21. Compound 21. Prepared as outlined for compound **23** in 85% yield. HPLC-MS (ESI^+): t_R=3.71 min, *m/z* 387 ($M+H^+$, 55%), 445 (12), 425 (8), 409 (29), 300 (6), 288 (24), 202 (78), 188 (18), 150 (100), 130 (7); δ_H (300 MHz, $CDCl_3$) 2.55 (t, 4H, J=4.2 Hz, CH_2N″CH_2), 2.66 (s, 3H, NCH_3), 2.85–3.25 (m, 8H, CH_2CH_2N′CH_2CH_2), 3.69 (t, 4H, J=4.2 Hz, CH_2OCH_2), 3.92 (s, 2H, CH_2N′), 3.95 (s, 3H, OMe), 4.81 (s, 2H, CH_2N″), 7.29 (s, 1H, H2), 7.47 (d, 1H, J=8.7 Hz, H8), 7.93 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.40 (d, 1H, J=1.5 Hz, H5); IR (film) 2951, 1707, 1614, 1115 cm^{-1}; HRMS (FAB+) calculated for $C_{21}H_{31}N_4O_3$ (M+H): 387.2396, found: 387.2392.

4.2.22. Compound 22. Prepared as outlined for compound **23** in 68% yield. HPLC-MS (ESI^+): t_R=4.32 min, *m/z* 443 ($M+Na^+$, 39%), 479 (7), 231 (6), 202 (77), 188 (100), 150 (52), 134 (100), 122 (36); δ_H (300 MHz, $CDCl_3$) 2.28 (s, 3H, N″CH_3), 2.65 (s, 3H, NCH_3), 2.85–3.25 (m, 8H, CH_2CH_2-N′CH_2CH_2), 3.62 (s, 2H, N″CH_2Ph), 3.92 (s, 2H, CH_2N′), 3.95 (s, 3H, OMe), 4.88 (s, 2H, CH_2N″), 7.22–7.36 (m, 6H, H2, Ar), 7.39 (d, 1H, J=8.7 Hz, H8), 7.92 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.37 (d, 1H, J=1.5 Hz, H5); IR (film) 2950, 1709, 1616, 1248 cm^{-1}; HRMS (FAB+) calculated for $C_{25}H_{33}N_4O_2$ (M+H): 421.2604, found: 421.2613.

4.2.23. Compound 24. Prepared as outlined for compound **23** in 57% yield. HPLC-MS (ESI^+): t_R=3.56 min, *m/z* 382 ($M+Na^+$, 100%), 418 (14), 404 (11), 297 (16), 287 (29), 202 (18), 188 (27), 158 (6), 144 (10), 137 (9), 120 (10); δ_H (300 MHz, $CDCl_3$) 1.08 (t, 6H, J=7.2 Hz, N″CH_2CH_3), 2.58 (m, 4H, CH_2N′CH_2), 2.61 (q, 4H, J=7.2 Hz, N″CH_2-CH_3), 3.75 (m, 4H, CH_2OCH_2), 3.80 (s, 2H, CH_2N′), 3.95 (s, 3H, OMe), 4.87 (s, 2H, CH_2N″), 7.27 (s, 1H, H2), 7.48 (d, 1H, J=8.7 Hz, H8), 7.89 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.48 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 12.6, 45.4, 51.2, 52.3, 53.6, 53.8, 67.0, 68.7, 110.0, 111.4, 122.1, 122.6, 122.9, 123.9, 127.9, 128.6, 139.3, 168.7; IR (film) 2951, 1709, 1619, 1115 cm^{-1}; HRMS (FAB+) calculated for $C_{20}H_{30}N_3O_3$ (M+H): 360.2287, found: 360.2282.

4.2.24. Compound 25. Prepared as outlined for compound **23** in 55% yield. HPLC-MS (ESI^+): t_R=3.79 min, *m/z* 409 ($M+Na^+$, 100%), 445 (20), 425 (6), 348 (7), 288 (8), 188 (100), 122 (7); δ_H (300 MHz, $CDCl_3$) 2.30 (s, 3H, NCH_3), 2.40–2.68 (m, 12H, CH_2N′CH_2, CH_2CH_2N″CH_2CH_2), 3.73 (m, 4H, CH_2OCH_2), 3.75 (s, 2H, CH_2N′), 3.94 (s, 3H, OMe), 4.80 (s, 2H, CH_2N″) 7.17 (s, 1H, H2), 7.43 (d, 1H, J=8.7 Hz, H8), 7.91 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.46 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 46.0, 50.4, 52.3, 53.8, 53.9, 55.0, 67.3, 68.4, 110.2, 122.0, 122.9, 123.7, 128.6, 129.9, 140.1, 168.5; IR (film) 2952, 1698, 1615, 1113 cm^{-1}; HRMS (FAB+) calculated for $C_{21}H_{31}N_4O_3$ (M+H): 387.2396, found: 387.2281.

4.2.25. Compound 26. Prepared as outlined for compound **23** in 51% yield. HPLC-MS (ESI): t_R=3.90 min, *m/z* 396 ($M+Na^+$, 100%), 432 (17), 412 (8), 287 (14), 188 (11), 144 (18); δ_H (300 MHz, $CDCl_3$) 2.53 (m, 4H, CH_2N″CH_2), 2.60 (m, 4H, CH_2N′CH_2), 3.68 (m, 4H, CH_2CH_2N″CH_2-CH_2), 3.76 (m, 4H, CH_2CH_2N′CH_2CH_2), 3.80 (s, 2H, CH_2N′), 3.95 (s, 3H, OMe), 4.79 (s, 2H, CH_2N″), 7.27 (s, 1H, H2), 7.45 (d, 1H, J=8.7 Hz, H8), 7.93 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.45 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 50.8, 52.0, 53.2, 66.7, 68.4, 109.9, 122.0, 122.3, 123.5, 128.2, 130.5, 139.6, 168.0; IR (film) 2923, 1710, 1614, 1115 cm^{-1}; HRMS (FAB+) calculated for $C_{20}H_{28}N_3O_4$ (M+H): 374.2080, found: 374.2081.

4.2.26. Compound 27. Prepared as outlined for compound **23** in 60% yield. HPLC-MS (ESI^+): t_R=4.52 min, *m/z* 430 ($M+Na^+$, 100%), 466 (13), 446 (9), 188 (94), 134 (74), 122 (8); δ_H (300 MHz, $CDCl_3$) 2.27 (s, 3H, N″CH_3), 2.61 (m, 4H, CH_2N′CH_2), 3.61 (s, 2H, N′CH_2Ph), 3.77 (m, 4H, CH_2OCH_2), 3.83 (s, 2H, CH_2N′), 3.94 (s, 3H, OMe), 4.86 (s, 2H, CH_2N″), 7.24–7.35 (m, 6H, H2, Ar), 7.38 (d, 1H, J= 8.7 Hz, H8), 7.91 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.45 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 40.6, 52.3, 53.5, 53.7, 59.4, 66.9, 67.7, 110.5, 122.2, 122.6, 123.7, 127.8, 128.5, 128.9, 129.1, 138.4, 140.1, 168.5; IR (film) 2923, 1709, 1614, 1254, 1116 cm^{-1}; HRMS (FAB+) calculated for $C_{24}H_{30}N_3O_3$ (M+H): 408.2287, found: 408.2300.

4.2.27. Compound 28. Prepared as outlined for compound **23** in 72% yield. HPLC-MS (ESI^+): t_R=4.65 min, *m/z* 442 ($M+Na^+$, 44%), 231 (100), 188 (7), 142 (20), 122 (13); δ_H (300 MHz, $CDCl_3$) 0.88 (d, 3H, J=5.7 Hz, $CHCH_3$), 1.23 (m, 3H, CHHCH(CH_3)CHH), 1.59 (m, 2H, CHHCH(CH_3)-CHH), 2.13 (m, 2H, CHHN″CHH), 2.24 (s, 3H, N′CH_3), 2.88 (m, 2H, CHHN″CHH), 3.60 (s, 2H, N′CH_2Ph), 3.77 (s, 2H, CH_2N′), 3.95 (s, 3H, OMe), 4.83 (s, 2H, CH_2N″), 7.15–7.47 (m, 7H, H2, H8, Ar), 7.90 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.46 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 22.2, 30.7, 34.5, 51.6, 52.2, 52.7, 62.2, 68.8, 110.2, 121.7, 123.0, 123.6, 127.6, 128.3, 128.7, 129.6, 140.4, 168.6; IR (film) 2924, 1716, 1614, 1245 cm^{-1}; HRMS (FAB+) calculated for $C_{26}H_{34}N_3O_2$ (M+H): 420.2651, found: 420.2640.

4.2.28. Compound 29. Prepared as outlined for compound **23** in 55% yield. HPLC-MS (ESI^+): t_R=4.42 min, *m/z* 416 ($M+Na^+$, 14%), 309 (8), 231 (28), 188 (40), 122 (100); δ_H (300 MHz, $CDCl_3$) 1.06 (t, 6H, J=7.2 Hz, N″CH_2CH_3), 2.24 (s, 3H, N′CH_3), 2.61 (q, 4H, J=7.2 Hz, N″CH_2CH_3), 3.61 (s, 2H, N′CH_2Ph), 3.79 (d, 2H, J=4.2 Hz, CH_2N′), 3.95 (s, 3H, OMe), 4.86 (s, 2H, CH_2N′), 7.18–7.44 (m, 7H, H2, H8, Ar), 7.90 (dd, 1H, J=8.7, 1.5 Hz, H7), 8.46 (d, 1H, J=1.5 Hz, H5); δ_C (75 MHz, DMSO) 21.0, 41.7, 51.6, 51.7, 58.3, 61.0, 66.8, 111.3, 122.4, 127.0, 127.4, 128.2, 128.5, 128.7, 136.1, 138.5, 139.7, 149.6, 167.3; IR (film) 2947, 1709, 1614, 1251 cm^{-1}; HRMS (FAB+) calculated for $C_{24}H_{32}N_3O_2$ (M+H): 394.2495, found: 394.2487.

4.2.29. Compound 30. Prepared as outlined for compound **23** in 63% yield. HPLC-MS (ESI^+): t_R=4.46 min, *m/z* 443 ($M+Na^+$, 67%), 459 (8), 277 (7), 188 (71), 180 (28), 134 (13), 122 (100); δ_H (300 MHz, $CDCl_3$) 2.24 (s, 3H, N′CH_3), 2.28 (s, 3H, NCH_3), 2.35–2.72 (m, 8H, CH_2CH_2N″CH_2-CH_2), 3.60 (s, 2H, N′CH_2Ph), 3.76 (s, 2H, CH_2N′), 3.95

(s, 3H, OMe), 4.79 (s, 2H, CH_2N''), 7.14–7.45 (m, 7H, H2, H8, Ar), 7.91 (dd, 1H, $J=8.7$, 1.5 Hz, H7), 8.46 (d, 1H, $J=$ 1.5 Hz, H5); δ_C (75 MHz, $CDCl_3$) 46.1, 50.5, 52.3, 52.6, 55.0, 62.1, 68.4, 110.2, 121.9, 123.0, 123.7, 127.5, 128.5, 128.7, 129.6, 140.2, 168.6; IR (film) 2947, 1709, 1614, 1251 cm^{-1}; HRMS (FAB+) calculated for $C_{25}H_{33}N_4O_2$ (M+H): 421.2604, found: 421.2609.

4.2.30. Compound 31. Prepared as outlined for compound **23** in 64% yield. HPLC-MS (ESI^+): $t_R=4.60$ min, *m/z* 430 ($M+Na^+$, 74%), 446 (10), 362 (20), 231 (100), 180 (8), 134 (6), 122 (14); δ_H (300 MHz, $CDCl_3$) 2.25 (s, 3H, $N'CH_3$), 2.52 (t, 4H, $J=4.5$ Hz, $CH_2N''CH_2$), 3.62 (s, 2H, $N'CH_2Ph$), 3.69 (t, 4H, $J=4.5$ Hz, CH_2OCH_2), 3.78 (s, 2H, CH_2N'), 3.95 (s, 3H, OMe), 4.78 (s, 2H, CH_2N''), 7.18–7.45 (m, 7H, H2, H8, Ar), 7.91 (dd, 1H, $J=8.7$, 1.5 Hz, H7), 8.46 (d, 1H, $J=1.5$ Hz, H5); δ_C (75 MHz, $CDCl_3$) 40.6, 52.3, 52.5, 59.4, 62.3, 67.6, 110.3, 121.8, 123.1, 123.7, 127.8, 128.5, 128.8, 128.9, 129.2, 138.4, 140.2, 168.6; IR (film) 2947, 1716, 1614, 1247 cm^{-1}.

4.2.31. Compound 32. Prepared as outlined for compound **23** in 56% yield. HPLC-MS (ESI^+): $t_R=5.10$ min, *m/z* 464 ($M+Na^+$, 56%), 480 (7), 231 (8), 188 (100), 134 (65), 122 (52); δ_H (300 MHz, $CDCl_3$) 2.26 (s, 6H, NCH_3), 3.61 (m, 4H, NCH_2Ph), 3.79 (s, 2H, CH_2N'), 3.95 (s, 3H, OMe), 4.86 (s, 2H, CH_2N''), 7.15–7.43 (m, 7H, H2, H8, Ar), 7.90 (dd, 1H, $J=8.7$, 1.5 Hz, H7), 8.47 (d, 1H, $J=1.5$ Hz, H5); δ_C (75 MHz, $CDCl_3$) 51.2, 52.3, 52.5, 67.1, 68.7, 110.2, 121.8, 123.0, 123.8, 128.5, 128.8, 129.7, 140.1, 168.5; IR (film) 2920, 1710, 1614, 1247, 1115 cm^{-1}; HRMS (FAB+) calculated for $C_{28}H_{32}N_3O_2$ (M+H): 444.2495, found: 444.2495.

4.2.32. Compound 33. To compound **2** (50.0 mg, 193.5 μmol) in dioxane–HOAc (4/1, 1.0 mL) was added a mixture of 4-methylpiperidine (34.3 μL, 290.2 μmol) and formaldehyde (21.7 μL, 290.2 μmol, 37 wt% solution in water) in dioxane–HOAc (4/1, 1.0 mL). The resultant mixture was agitated at room temperature for 66 h. Evaporation of the solvents and flash chromatography (CH_2Cl_2/MeOH/Et_3N, v/v/v 90:10:1) yielded **33** (46 mg, 64%). HPLC-MS (ESI^+): $t_R=4.12$ min, *m/z* 370 ($M+H^+$); δ_H (300 MHz, $CDCl_3$) 0.87 (d, 3H, $J=5.4$ Hz, H-21), 1.21 (m, 3H, H-19, H18, H17), 1.60 (m, 2H, H19, H17), 1.88 (m, 4H, H13, H12), 2.13 (m, 2H, H20, H16), 2.81 (m, 4H, H14, H11), 2.88 (m, 2H, H-20, H-16), 3.94 (s, 3H, OMe), 4.01 (s, 2H, H10), 4.84 (s, 2H, H15), 7.38 (s, 1H, H2), 7.49 (d, 1H, $J=8.7$ Hz, H8), 7.91 (dd, 1H, $J=8.7$, 1.6 Hz, H7), 8.39 (d, 1H, $J=1.6$ Hz, H5); δ_C (75 MHz, $CDCl_3$) 22.2 (C21), 23.8 (C13, C12), 30.7 (C18), 34.4 (C19, C17), 50.0 (C20), 51.6 (C16), 52.3 (C10), 54.1 (C11, C14), 68.9 (C15), 110.5 (C8), 111.8 (C3), 121.8 (C6), 122.1 (C5), 123.6 (C7), 128.1 (C4), 130.9 (C2), 140.0 (C9), 168.5 (CO).

4.2.33. Compound 34. To compound **3** (104 mg, 362 μmol) in dioxane–HOAc (4/1, 1.0 mL) was added a mixture of pyrrolidine (45 μL, 543 μmol) and formaldehyde (41 μL, 543 μmmol, 37 wt% solution in water) in dioxane–HOAc (4/1, 1.0 mL). After agitation of the resultant mixture for 48 h at room temperature, an additional portion of pyrrolidine (90 μL, 1.09 mmol) and formaldehyde (82 μL, 1.09 mmol, 37 wt% solution in water). After agitation for an additional 18 h at the solvents were removed and the residue was flash chromatographed (CH_2Cl_2/MeOH/Et_3N, v/v/v 90:10:1) to give **34** (83 mg, 62%). δ_H (300 MHz, $CDCl_3$) 0.91 (d, 3H, $J=5.7$ Hz, H21), 1.30 (m, 3H, H19, H18, H17), 1.60 (m, 2H, H19, H17), 1.75 (m, 4H, H13, H12), 2.03 (m, 2H, H20, H16), 2.64 (m, 4H, H14, H11), 2.96 (m, 2H, H20, H16), 3.75 (s, 2H, H15), 3.93 (s, 3H, OMe), 4.96 (s, 2H, H10), 7.23 (s, 1H, H2), 7.44 (d, 1H, $J=8.7$ Hz, H8), 7.91 (dd, 1H, $J=8.7$, 1.5 Hz, H7), 8.43 (d, 1H, $J=1.5$ Hz, H5); δ_C (75 MHz, $CDCl_3$) 22.2 (C21), 24.0 (C12, C13), 31.0 (C18), 34.4 (C17, C19), 51.4 (C14, C11), 52.2 (MeO), 53.4 (C15), 54.0 (C20, C16), 64.9 (C10), 110.0 (C8), 112.7 (C3), 121.8 (C6), 122.6 (C5), 123.4 (C7), 128.7 (C4), 130.3 (C2), 139.9 (C9), 168.6 (CO).

Acknowledgements

This work was supported financially by Amersham Biosciences and the Royal Institute of Technology. We also wish to thank Dr. Herbert Baumann, Amersham Biosciences, for assistance with the NMR experiments.

References and notes

1. (a) Wilchek, M.; Chaiken, I. *Methods Mol. Biol.* **2000**, *147*, 1–6. (b) Scopes, R. K. *Protein Purification; Principles and Practice*; Springer: New York, 1994.
2. Dolle, R. E. *J. Comb. Chem.* **2004**, *6*, 623–679.
3. (a) Wu, T. Y. H.; Ding, S.; Gray, N.; Schultz, P. G. *Org. Lett.* **2001**, *3*, 3827–3830. (b) Zhang, H.-C.; Derian, C. K.; Andrade-Gordon, P.; Hoekstra, W. J.; McComsey, D. F.; White, K. B.; Poulter, B. L.; Addo, M. F.; Cheung, W.-M.; Damiano, B. P.; Oksenberg, D.; Reynolds, E. E.; Pandey, A.; Scarborough, R. M.; Maryanoff, B. E. *J. Med. Chem.* **2001**, *44*, 1021–1024. (c) Zhang, H.-C.; McComsey, D. F.; White, K. B.; Addo, M. F.; Andrade-Gordon, P.; Derian, C. K.; Oksenberg, D.; Maryanoff, B. E. *Bioorg. Med. Chem. Lett.* **2001**, *11*, 2105–2109.
4. Joule, J. A.; Mills, K.; Smith, G. F. *Heterocyclic Chemistry*; Chapman: London, 1995.
5. (a) Abel, E.; De Wall, S. L.; Edwards, W. B.; Lalitha, S.; Covey, D. F.; Gokel, G. W. *J. Org. Chem.* **2000**, *65*, 5901–5909. (b) Reinecke, M. G.; Sebastian, J. F.; Johnson, H. W.; Pyun, C. *J. Org. Chem.* **1972**, *37*, 3066–3068.
6. (a) Zhang, H.-C.; Brumfield, K. K.; Jaroskova, L.; Maryanoff, B. E. *Tetrahedron Lett.* **1998**, *39*, 4449–4452. (b) Dauzonne, D.; O'Neil, I. A.; Renaud, A. *J. Org. Chem.* **1984**, *49*, 4409–4415.
7. Katritzky, A. R.; Lue, P.; Chen, Y.-X. *J. Org. Chem.* **1990**, *55*, 3688–3691.
8. Bax, A.; Griffey, R. H.; Hawkins, B. L. *J. Magn. Reson.* **1983**, *55*, 301–315.
9. Bax, A.; Summers, M. F. *J. Am. Chem. Soc.* **1986**, *108*, 2093–2094.
10. The HMBC was performed with 16 scans and 256 increments using long-range coupling evolution time of 65 ms.
11. Booth, R. J.; Hodges, J. C. *J. Am. Chem. Soc.* **1997**, *119*, 4882–4886.