Synthesis and Cytotoxic Activity of Dimeric Analogs of Acronycine in the Benzo[*b*]pyrano[3,2-*h*]acridin-7-one Series

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Coupling of 6-hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (6) with α,ω-diiodoalkanes of varying length under alkaline conditions gave dimers 7—10. Halogenated ethers 11—14, cyclization products 15—17, and compounds 18—22 were also isolated in small yield from the reaction mixtures. Compounds 7—10 were more potent than acronycine and benzo[*b*]acronycine in inhibiting L1210 cell proliferation. The length of the alkyl ether linkage between the two benzopyranoacridone units had a dramatic influence on the cytotoxic activity. Compound 9 ($n=5$) was the most active, with an IC_{50} value against L1210 cells within the same range of magnitude as diacetate 5, currently under clinical development.

Key words benzo[*b*]pyrano[3,2-*h*]acridin-7-one dimer; acronycine; synthesis; cytotoxicity

From a biogenetic point of view, homo and hetero dimeric secondary metabolites present in plants and microorganisms generally arise from oxidative coupling and/or pericyclic reactions.[1—4] In the field of alkaloids, such compounds often exhibit biological activities dramatically increased, when compared with the corresponding monomeric units, as illustrated by anticancer heterodimeric bisindole alkaloids vinblastine and vincristine,[5] and curarizing homodimeric bisindole and bisisoquinoline alkaloids toxiferine and tubocurarine.[6] As far as DNA interacting agents are concerned, the idea of mimicking Nature led to conceive dimeric synthetic systems, which also display significantly enhanced biological activity when compared to the parent monomeric entities. Ditercalinium (**1**), resulting from the dimerization of two pyridocarbazole units related to natural alkaloids ellipticine (**2**) and olivacine (**3**) is an excellent example of this approach.[7—9]

Benzo[*b*]acronycine derived antitumor derivatives, developed from the model of natural acronycine (**4**),[10—14] are exemplified by diacetate **5**, currently under phase I clinical trials under the code S23906-1.[15,16] These compounds displayed a particularly impressive broad antitumor spectrum, when evaluated against aggressive orthotopic models of human ovarian, lung, and colon cancers. The mechanism of action of these compounds has been recently shown to imply alkylation of the 2-amino group of DNA guanine residues.[17—19] In this context, benzo[*b*]acronycine dimers appeared us as possible drug candidates. The linkage of the two monomeric units was envisaged through their C-6 positions, since acronycine and benzo[*b*]acronycine derivatives modified at this position had been previously shown to exhibit cytotoxic activities *in vitro* and antitumor properties *in vivo* comparable or superior to those of the parent compounds.[20,21]

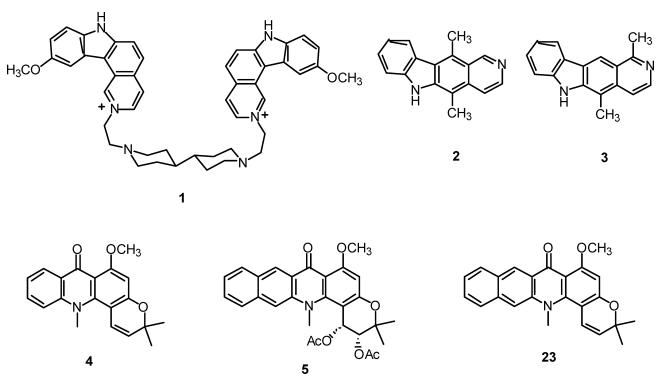

Fig. 1

Chemistry A flexible α,ω-alkyl ether linkage was envisaged to tether the two benzo[*b*]acronycine units, since the use of this type of spacer recently resulted in dimers with significantly enhanced DNA-binding affinities when compared to the corresponding monomers in the cytotoxic pyrrolo[2,1-*c*][1,4]benzodiazepine[22—24] and berberine[25] series.

Coupling of 6-hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**6**)[13] with α,ω-diiodoalkanes of varying length under alkaline conditions, according to a method essentially similar to that initially introduced by Marvel and Tannenbaum,[26] provided the desired dimers **7**—**10** in 10—32% yield. Halogenated ethers **11**—**14**, arising from monoalkylation at O-6, were also isolated in 9—34% yield from the reaction mixtures. They were accompanied by minute amounts of intramolecular cyclization products at C-5, illustrated by the fused pyran **15**, oxepine **16**, and oxocine **17**, when 1,3-diiodopropane, 1,4-diiodobutane, and 1,5-diiodopentane were used as alkylating agents, respectively.[27] In addition, compounds resulting from *C*-alkylation at C-5 and olefins obtained through hydrogen iodide elimination, **18**—**22**, were obtained in small yield, particularly in the course of the reaction involving 1,3-diiodopropane.

Pharmacology The study of the cytotoxic properties of the benzo[*b*]acronycine dimers **7**—**10** and of halogenated ethers **11**—**14** was carried out *in vitro* on the L1210 murine leukemia cell line. The results (IC_{50}) are reported in Table 1, in comparison with acronycine (**4**), benzo[*b*]acronycine (**23**), and 6-hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-

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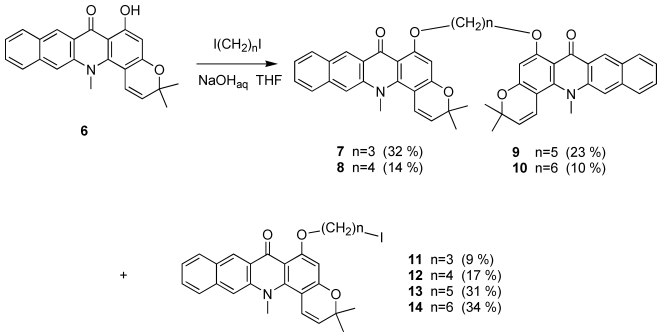
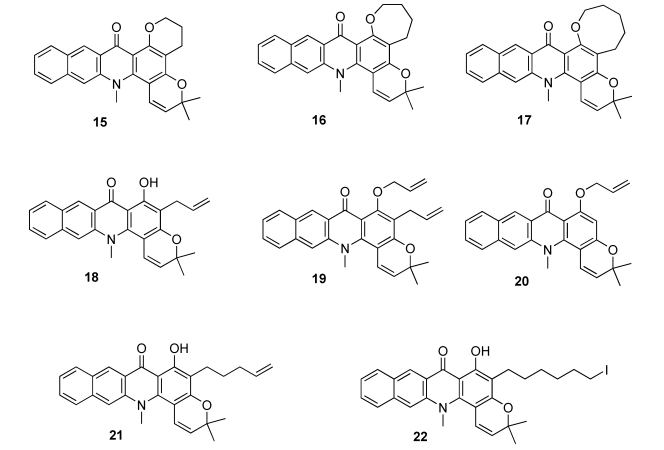


Chart 1. Synthesis of Dimeric Analogs of Acronycine

Table 1. Inhibition of L1210 Cell Proliferation and Cell Cycle Perturbation Induced by Dimeric Compounds **7**—**10** and Halogenated Ethers **11**—**14**, in Comparison with Acronycine (**4**), Benzo[*b*]acronycine (**23**), (±)-*cis*-1,2-Diacetoxy-1,2-dihydrobenzo[*b*]acronycine (**5**), and 6-Hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**6**)

Compound	Cytotoxicity IC_{50} (μM)
7	7.2
8	5.9
9	0.9
10	5.3
11	2.0
12	5.8
13	3.9
14	4.1
4	23.2
23	14.9
5	0.7
6	17.9



pyrano[3,2-*h*]acridin-7-one (**6**). As expected, dimeric compounds **7**—**10** were more potent than parent acronycine and benzo[*b*]acronycine in inhibiting L1210 cell proliferation. Interestingly, introduction of a iodoalkylether side chain at position 6 in compounds **11**—**14** also resulted significant increase of the cytotoxic activity when compared with the parent compounds bearing a methoxy or a hydroxy group at C-6.

The perturbation of the cell cycle induced by the most active benzo[*b*]acronycine dimer **9** was studied on L1210 cell line. As previously observed for acronycine itself,[12] compound **9** induced a partial accumulation of cells in the G_2+M phase at 2.5 μM (32% accumulation in treated cells *versus* 21% in control cells). Induction of apoptosis was observed after 24 h of exposure at 5 μM.[28]

Results and Discussion

In terms of structure activity relationships, the length of the alkyl ether linkage between the two benzopyranoacridone units appears as a particularly important feature in this dimeric series. Indeed, compounds **7** (n=3), **8** (n=4) and **10** (n=6) only exhibited a moderate increase of the cytotoxic activity when compared to the parent compounds. In contrast, compound **9** (n=5) was markedly more potent in inhibiting L1210 cell proliferation, with a IC_{50} value within the same range of magnitude as diacetate **5**, currently under clinical development.

Experimental

Chemistry Mass spectra were recorded with a Waters ZQ 2000 spectrometer using electrospray ionization (ES-MS). UV spectra (λ_{max} in nm) were recorded in spectroscopic grade MeOH on a Beckman Model 34 spectrophotometer. IR spectra (ν_{max} in cm^{-1}) were obtained from potassium bromide pellets or sodium chloride films on a Perkin-Elmer 257 instrument. ^{1}H-NMR (δ[ppm], J[Hz]) spectra were run at 400 MHz and ^{13}C-NMR spectra at 100 MHz, using a Bruker AVANCE-400 spectrometer. When necessary, the structures of the novel compounds were insured and the signals unambiguously assigned by 2D NMR techniques: ^{1}H–^{1}H COSY, ^{1}H–^{1}H NOESY, ^{13}C–^{1}H HMQC, and ^{13}C–^{1}H HMBC. These experiments were performed using standard Bruker microprograms. Column chromatographies were conducted using silica gel 60 Merck (20—45 μm) with an overpressure of 300 mbar.

Reaction of 6-Hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (6) with 1,3-Diiodopropane A 12 M aqueous sodium hydroxide solution 50 μl, (0.6 mmol) was added to a solution of **6** (214 mg, 0.6 mmol) and 1,3-diiodopropane (210 μl, 1.8 mmol) in tetrahydrofuran (10 ml). The resulting mixture was refluxed for 23 h. The solvents were evaporated under reduced pressure. The solid residue was extracted with CH_2Cl_2 (3×30 ml). The organic layers were dried over anhydrous $MgSO_4$, filtered and evaporated under reduced pressure. Column chromatography over silica gel (eluent: CH_2Cl_2 then CH_2Cl_2/MeOH mixtures of increasing polarity) gave successively 6-hydroxy-5-(prop-2-enyl)-3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**18**) (10 mg, 4%), 5-(prop-2-enyl)-6-(prop-2-enyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**19**) (10 mg, 4%), 6-(3-iodopropyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**11**) (29 mg, 9%), 6-(prop-2-enyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**20**) (22 mg, 9%), 6,6,9-trimethyl-3,4,6,9-tetrahydro-2*H*,16*H*-benzo[*i*]dipyrano[2,3-*a*:2′,3′-*c*]acridin-16-one (**15**) (29 mg, 13%), and 6,6′-(propane-1,3-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one) (**7**) (68 mg, 32%).

6-Hydroxy-5-(prop-2-enyl)-3,3,14-trimethyl-3,14-dihydro-7*H*-benzo-[*b*]pyrano[3,2-*h*]acridin-7-one (18) Amorphous solid, UV λ (MeOH) (log ε) 209 (4.46), 250 (4.39), 281 (4.71), 301 (4.77), 313 (4.77), 374 (4.06), 471 (3.70) nm; IR (KBr) ν 3435, 3081, 3052, 2973, 2923, 2854, 1636, 1624, 1583, 1552, 1490, 1467, 1451, 1409, 1374, 1331, 1184, 1137, 1121, 896, 864 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.55 (6H, s, CMe_2), 3.44 (2H, d, J=6, CH_2-1′), 3.95 (3H, s, NMe), 4.99 (1H, dd, J=10, 2, H-3′a), 5.12 (1H, dd, J=17, 2, H-3′b), 5.55 (1H, d, J=10, H-2), 6.01 (1H, ddt, J=17, 10, 6, H-2′), 6.60 (1H, d, J=10, H-1), 7.42 (1H, t, J=8, H-10), 7.57 (1H, t, J=8, H-11), 7.69 (1H, s, H-13), 7.87 (1H, d, J=8, H-12), 7.98 (1H, d, J=8, H-9), 8.94 (1H, s, H-8), 14.90 (1H, s, OH); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 26.7 (CH_2-1′), 27.1 ($C(\underline{C}H_3)_2$), 44.2 ($N\underline{C}H_3$), 76.5 (C-3), 100.9 (C-14b), 105.8 (C-6a), 107.7 (C-5), 112.2 (C-13), 114.5 (C-3′), 122.1 (C-1), 122.2 (C-7a), 122.9 (C-2), 124.9 (C-10), 127.0 (C-12), 127.7 (C-8), 128.4 (C-8a), 128.9 (C-11), 129.6 (C-9), 136.4 (C-12a), 136.5 (C-2′), 141.9 (C-13a), 143.7 (C-14a), 160.2 (C-4a), 162.9 (C-6), 182.2 (C-7); ES-MS m/z 420 $[MNa]^+$, 398 $[MH]^+$. *Anal.* Calcd for $C_{26}H_{23}NO_3$: C, 78.57; H, 5.83; N, 3.52. Found: C, 78.65; H, 5.79; N, 3.58.

5-(Prop-2-enyl)-6-(prop-2-enyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (19) Amorphous solid, ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.56 (6H, s, CMe_2), 3.50 (2H, d, J=6, CH_2-1′), 3.93

(3H, s, NMe), 4.60 (2H, d, J=6, CH_2-1″), 4.99 (1H, dd, J=10, 1, H-3′a), 5.08 (1H, dd, J=16, 1, H-3′b), 5.31 (1H, dd, J=10, 1, H-3″a), 5.50 (1H, dd, J=17, 1, H-3″b), 5.61 (1H, d, J=10, H-2), 6.00 (1H, ddt, J=16, 10, 6, H-2″), 6.32 (1H, m, H-2″), 6.62 (1H, d, J=10, H-1), 7.42 (1H, t, J=8, H-10), 7.54 (1H, t, J=8, H-11), 7.69 (1H, s, H-13), 7.88 (1H, d, J=8, H-12), 8.03 (1H, d, 1H, J=8, H-9), 8.98 (1H, s, H-8); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 26.7 (C($\underline{C}H_3$)$_2$), 27.7 (C-1′), 44.6 (N$\underline{C}H_3$), 75.3 (C-1″), 76.0 (C-3), 106.3 (C-14b), 111.6 (C-13), 112.2 (C-6a), 114.6 (C-3′), 116.5 (C-5), 117.0 (C-3″), 122.0 (C-1), 124.1 (2C, C-2, C-10), 124.8 (C-7a), 126.6 (C-12), 128.3 (C-8), 128.6 (C-8a), 129.4 (C-9), 130.7 (C-11), 134.4 (C-2″), 135.7 (C-12a), 136.9 (C-2′), 141.8 (C-13a), 145.6 (C-14a), 157.8 (C-4a), 158.7 (C-6), 177.4 (C-7); ES-MS m/z 476 $[MK]^+$, 460 $[MNa]^+$, 438 $[MH]^+$. *Anal.* Calcd for $C_{29}H_{27}NO_3$: C, 79.61; H, 6.22; N, 3.20. Found: C, 79.75; H, 6.19; N, 3.12.

6-(3-Iodopropyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (11) Amorphous solid, UV λ (MeOH) (log ε) 206 (4.51), 240 (4.34), 276 (4.62), 298 (4.62), 308 (4.65), 364 (3.88), 445 (3.69) nm; IR (KBr) ν 3048, 3017, 2959, 2921, 2854, 1646, 1631, 1614, 1588, 1561, 1495, 1460, 1398, 1355, 1332, 1188, 1137, 1087, 803 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.57 (6H, s, CMe_2), 2.43 (2H, quintet, J=6, CH_2-2′), 3.72 (2H, t, J=6, CH_2-3′), 3.92 (3H, s, NMe), 4.19 (2H, t, J=6, CH_2-1′), 5.54 (1H, d, J=10, H-2), 6.31 (1H, s, H-5), 6.59 (1H, d, J=10, H-1), 7.42 (1H, t, J=8, H-10), 7.55 (1H, t, J=8, H-11), 7.67 (1H, s, H-13), 7.87 (1H, d, J=8, H-12), 8.01 (1H, d, 1H, J=8, H-9), 8.93 (1H, s, H-8); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 4.4 (C-3′), 27.0 (C($\underline{C}H_3$)$_2$), 32.8 (C-2′), 44.9 (N$\underline{C}H_3$), 68.2 (C-1′), 76.6 (C-3), 94.9 (C-5), 103.3 (C-14b), 109.7 (C-6a), 112.0 (C-13), 122.1 (C-1), 123.2 (C-2), 124.6 (C-10), 125.4 (C-7a), 126.8 (C-12), 128.2 (C-8), 128.3 (C-11), 128.7 (C-8a), 129.7 (C-9), 135.8 (C-12a), 142.1 (C-13a), 147.7 (C-14a), 159.9 (C-4a), 162.4 (C-6), 178.0 (C-7); ES-MS m/z 548 $[MNa]^+$, 526 $[MH]^+$. *Anal.* Calcd for $C_{26}H_{24}INO_3$: C, 59.44; H, 4.60; N, 2.67; I, 24.15. Found: C, 59.53; H, 4.69; N, 2.61; I, 24.25.

6-(Prop-2-enyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (20) Amorphous solid, UV λ (MeOH) (log ε) 206 (4.54), 241 (4.38), 275 (4.66), 299 (4.66), 308 (4.69), 363 (3.90), 445 (3.71) nm; IR (KBr) ν 3052, 2974, 2966, 2924, 2846, 1650, 1619, 1584, 1561, 1491, 1456, 1401, 1359, 1332, 1172, 1141, 1118, 1095, 982, 920, 846 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.56 (6H, s, CMe_2), 3.92 (3H, s, NMe), 4.71 (2H, d, J=5 Hz, CH_2-1′), 5.40 (1H, dd, J=10, 2, H-3′a), 5.54 (1H, d, J=10, H-2), 5.78 (1H, dd, J=17, 2, H-3′b), 6.19 (1H, ddt, J=17, 10, 5, H-2′), 6.29 (1H, s, H-5), 6.59 (1H, d, J=10, H-1), 7.41 (1H, t, J=8, H-10), 7.54 (1H, t, J=8, H-11), 7.67 (1H, s, H-13), 7.87 (1H, d, J=8, H-12), 8.01 (1H, d, J=8, H-9), 8.95 (1H, s, H-8); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 27.0 (C($\underline{C}H_3$)$_2$), 44.9 (N$\underline{C}H_3$), 69.7 (C-1′), 76.5 (C-3), 95.2 (C-5), 103.3 (C-14b), 109.9 (C-6a), 111.9 (C-13), 117.9 (C-3′), 122.1 (C-1), 123.2 (C-2), 124.5 (C-10), 125.6 (C-7a), 126.8 (C-12), 128.3 (C-11), 128.4 (C-8), 128.7 (C-12a), 129.7 (C-9), 132.5 (C-2′), 135.8 (C-8a), 142.1 (C-13a), 147.7 (C-14a), 159.7 (C-4a), 162.2 (C-6), 178.0 (C-7); ES-MS m/z 420 $[MNa]^+$, 398 $[MH]^+$. *Anal.* Calcd for $C_{26}H_{23}NO_3$: C, 78.57; H, 5.83; N, 3.52. Found: C, 78.66; H, 5.83; N, 3.48.

6,6,9-Trimethyl-3,4,6,9-tetrahydro-2*H*,16*H*-benzo[*i*]dipyrano[2,3-*a*:2′,3′-*c*]acridin-16-one (15) Amorphous solid, UV λ (MeOH) (log ε) 208 (4.46), 243 (4.36), 280 (4.65), 299 (4.61), 311 (4.61), 367 (3.88), 452 (3.67) nm; IR (KBr) ν 3048, 2962, 2924, 2850, 1650, 1631, 1615, 1579, 1487, 1456, 1440, 1413, 1356, 1343, 1242, 1184, 1137, 1122, 1114, 1067, 1029, 840 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.55 (6H, s, CMe_2), 2.06 (2H, m, CH_2-3), 2.69 (2H, t, J=7, CH_2-4), 3.88 (3H, s, NMe), 4.41 (2H, t, J=5, CH_2-2), 5.52 (1H, d, J=10, H-7), 6.58 (1H, d, J=10, H-8), 7.39 (1H, t, J=8, H-13), 7.52 (1H, t, J=8, H-12), 7.63 (1H, s, H-10), 7.84 (1H, d, J=8, H-11), 8.00 (1H, d, J=8, H-14), 8.93 (1H, s, H-15); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 19.3 (C-4), 20.9 (C-3), 27.2 (C($\underline{C}H_3$)$_2$), 44.9 (N$\underline{C}H_3$), 67.3 (C-2), 76.4 (C-6), 102.4 (C-8a), 104.8 (C-4a), 109.3 (C-16a), 111.7 (C-10), 122.4 (C-8), 122.8 (C-7), 124.3 (C-13), 125.8 (C-15a), 126.8 (C-11), 128.1 (C-12), 128.2 (C-15), 128.6 (C-14a), 129.7 (C-14), 135.8 (C-10a), 142.3 (C-9a), 145.6 (C-8b), 157.4 (C-4b), 158.4 (C-16b), 178.2 (C-16); ES-MS m/z 420 $[MNa]^+$, 398 $[MH]^+$. *Anal.* Calcd for $C_{26}H_{23}NO_3$: C, 78.57; H, 5.83; N, 3.52. Found: C, 78.49; H, 5.91; N, 3.57.

6,6′-(Propane-1,3-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one) (7) Amorphous solid, UV λ (MeOH) (log ε) 206 (4.75), 241 (4.61), 277 (4.86), 298 (4.83), 307 (4.83), 363 (4.08), 439 (3.91) nm; IR (KBr) ν 3052, 2970, 2924, 2850, 1650, 1619, 1588, 1557, 1491, 1460, 1398, 1359, 1328, 1234, 1184, 1141, 1122, 1083, 815 cm^{-1}; ^{1}H-NMR (400 MHz, DMSO-d_6) δ 1.44 (12H, s, 2 CMe_2), 2.30 (2H, m, CH_2-2″), 3.84 (6H, s, 2NMe), 4.46 (4H, t, J=5, CH_2-1″, CH_2-3″), 5.59 (2H, d, J=10, H-2, H-2′), 6.45 (2H, s, H-5, H-5′), 6.68 (2H, d, J=10, H-1, H-1′), 7.44 (2H, t, J=8, H-10, H-10′), 7.59 (2H, t, J=8, H-11, H-11′), 7.91 (2H, s, H-13, H-13′), 7.99 (2H, d, J=8, H-12, H-12′), 8.07 (2H, d, J=8, H-9, H-9′), 8.72 (2H, s, H-8, H-8′); ES-MS m/z 777 $[MNa]^+$, 755 $[MH]^+$. *Anal.* Calcd for $C_{49}H_{42}N_2O_6$: C, 77.96; H, 5.61; N, 3.71. Found: C, 78.05; H, 5.68; N, 3.66.

Reaction of 6-Hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (6) with 1,4-Diiodobutane Alkylation of **6** (214 mg, 0.6 mmol) with 1,4-diiodobutane (230 ml, 1.8 mmol) was performed according to the procedure described for the condensation of **6** with 1,3-diiodopropane. Column chromatography over silica gel (eluent: CH_2Cl_2 then CH_2Cl_2/MeOH mixtures of increasing polarity) successively afforded 6-(4-iodobutyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**12**) (55 mg, 17%), 7,7,10-trimethyl-2,3,4,5,7,10-hexahydro-17*H*-benzo[*i*]oxepino[2,3-*a*]pyrano[2,3-*c*]acridin-17-one (**16**) (8 mg, 3%), and 6,6′-(butane-1,4-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one) (**8**) (33 mg, 14%).

6-(4-Iodobutyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (12) Amorphous solid, UV λ (MeOH) (log ε) 206 (4.56), 241 (4.40), 276 (4.68), 298 (4.67), 308 (4.70), 363 (3.91), 444 (3.73) nm; IR (KBr) ν 3040, 3013, 2970, 2920, 2867, 1646, 1615, 1588, 1565, 1491, 1398, 1351, 1335, 1223, 1188, 1137, 1083, 858, 803 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.56 (6H, s, CMe_2), 2.10 (2H, m, CH_2-2′), 2.25 (2H, m, CH_2-3′), 3.38 (2H, t, J=7, CH_2-4′), 3.89 (3H, s, NMe), 4.13 (2H, t, J=6, CH_2-1′), 5.53 (1H, d, J=10, H-2), 6.27 (1H, s, H-5), 6.57 (1H, d, J=10, H-1), 7.40 (1H, t, J=8, H-10), 7.53 (1H, t, J=8, H-11), 7.64 (1H, s, H-13), 7.85 (1H, d, J=8, H-12), 7.99 (1H, d, J=8, H-9), 8.93 (1H, s, H-8); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 7.6 (C-4′), 27.0 (C($\underline{C}H_3$)$_2$), 29.9 (2C, C-2′, C-3′), 44.9 (N$\underline{C}H_3$), 67.9 (C-1′), 76.6 (C-3), 94.7 (C-5), 103.2 (C-14b), 109.7 (C-6a), 112.0 (C-13), 122.1 (C-1), 123.1 (C-2), 124.6 (C-10), 125.5 (C-7a), 126.8 (C-12), 128.2 (2C, C-8, C-11), 128.7 (C-8a), 129.7 (C-9), 135.8 (C-12a), 142.1 (C-13a), 147.7 (C-14a), 159.8 (C-4a), 162.6 (C-6), 178.0 (C-7); ES-MS m/z 562 $[MNa]^+$, 540 $[MH]^+$. *Anal.* Calcd for $C_{27}H_{26}INO_3$: C, 60.12; H, 4.86; N, 2.60; I, 23.53. Found: C, 60.02; H, 4.79; N, 2.56; I, 23.62.

7,7,10-Trimethyl-2,3,4,5,7,10-hexahydro-17*H*-benzo[*i*]oxepino[2,3-*a*]pyrano[2,3-*c*]acridin-17-one (16) Amorphous solid, UV λ (MeOH) (log ε) 207 (4.35), 276 (4.43), 311 (4.31), 446 (3.41) nm; IR (KBr) ν 3052, 2920, 2854, 1619, 1574, 1489, 1456, 1401, 1354, 1285, 1180, 1126, 1104, 1040, 730 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.56 (6H, s, CMe_2), 1.74 (2H, m, CH_2-4), 2.07 (2H, m, CH_2-3), 2.94 (2H, m, CH_2-5), 3.90 (3H, s, NMe), 4.26 (2H, m, CH_2-2), 5.59 (1H, d, J=10, H-8), 6.62 (1H, d, J=10, H-9), 7.41 (1H, t, J=8, H-14), 7.52 (1H, t, J=8, H-13), 7.63 (1H, s, H-11), 7.84 (1H, d, J=8, H-12), 8.00 (1H, d, J=8, H-15), 8.93 (1H, s, H-16); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 23.8 (C-5), 25.4 (C-4), 26.9 (C($\underline{C}H_3$)$_2$), 29.7 (C-3), 44.7 (N$\underline{C}H_3$), 73.5 (C-2), 76.1 (C-7), 105.8 (C-9a), 111.8 (C-11), 112.3 (C-17a), 119.3 (C-5a), 122.4 (C-9), 123.9 (C-8), 124.3 (C-14), 125.5 (C-16a), 126.7 (C-12), 128.2 (2C, C13, C-16), 128.5 (C-15a), 129.7 (C-15), 135.8 (C-11a), 142.4 (C-10a), 144.8 (C-9b), 156.7 (C-5b), 163.2 (C-17b), 178.4 (C-17); ES-MS m/z 434 $[MNa]^+$, 412 $[MH]^+$. *Anal.* Calcd for $C_{27}H_{25}NO_3$: C, 78.81; H, 6.12; N, 3.40. Found: C, 78.74; H, 6.05; N, 3.44.

6,6′-(Butane-1,4-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one) (8) Amorphous solid, IR (KBr) ν 3044, 3013, 2975, 2970, 2924, 2866, 1646, 1619, 1588, 1557, 1491, 1460, 1394, 1355, 1335, 1238, 1184, 1141, 1083, 862, 827, 807, 755, 745 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$/CD_3OD 9/1 v/v) δ 1.47 (12H, s, 2 CMe_2), 2.36 (4H, m, CH_2-2″, CH_2-3″), 3.88 (6H, s, 2 NMe), 4.28 (m, 4H, CH_2-1″, CH_2-4″), 5.46 (2H, d, J=10, H-2, H-2′), 6.33 (2H, s, H-5, H-5′), 6.52 (2H, d, J=10, H-1, H-1′), 7.39 (2H, t, J=8, H-10, H-10′), 7.53 (2H, t, J=8, H-11, H-11′), 7.64 (2H, s, H-13, H-13′), 7.85 (2H, d, J=8, H-12, H-12′), 8.00 (2H, d, J=8, H-9, H-9′), 8.92 (2H, s, H-8, H-8′); ^{13}C-NMR (100 MHz, $CDCl_3$/CD_3OD 9/1 v/v) δ 25.8 (C-2″, C-3″); 26.8 (2 C($\underline{C}H_3$)$_2$), 44.7 (2 N$\underline{C}H_3$), 68.8 (C-1″, C-4″), 76.2 (C-3, C-3′), 94.9 (C-5, C-5′), 102.8 (C-14b, C-14b′), 109.4 (C-6a, C-6a′), 111.8 (C-13, C-13′), 121.9 (C-1, C-1′), 122.9 (C-2, C-2′), 124.3 (C-10, C-10′), 125.4 (C-7a, C-7a′), 126.6 (C-12, C-12′), 128.1 (C-8, C-8′, C-11, C-11′), 128.6 (C-12a, C-12a′), 129.6 (C-9, C-9′), 135.7 (C-8a, C-8a′), 142.0 (C-13a, C-13a′), 147.5 (C-14a, C-14a′), 159.9 (C-4a, C-4a′), 162.8 (C-6, C-6′), 178.1 (C-7, C-7′); ES-MS m/z 791 $[MNa]^+$, 769 $[MH]^+$. *Anal.* Calcd for $C_{50}H_{44}N_2O_6$: C, 78.10; H, 5.77; N, 3.64. Found: C, 78.03; H, 5.84; N, 3.71.

Reaction of 6-Hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (6) with 1,4-Diiodopentane Alkylation of **6** (1.00 g, 2.8 mmol) with 1,5-diiodopentane (1.04 ml, 7.0 mmol) was performed according to the procedure described for the condensation of **6** with 1,3-diiodopropane. Column chromatography over silica gel (eluent: CH_2Cl_2 then CH_2Cl_2/MeOH mixtures of increasing polarity) successively

afforded 6-hydroxy-5-(pent-4-enyl)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**21**) (38 mg, 4%), 6-(5-iodopentyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**13**) (475 mg, 31%), 8,8,11-trimethyl-3,4,5,6,8,11-2*H*,18*H*-benzo[*i*]oxocino[2,3-*a*]pyrano[2,3-*c*]acridin-18-one (**17**) (10 mg, 1%), and 6,6′-(pentane-1,5-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one) (**9**) (254 mg, 23%).

6-Hydroxy-5-(pent-4-enyl)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (21) Amorphous solid, UV λ (MeOH) (log ε) 206 (4.42), 250 (4.35), 281 (4.67), 301 (4.73), 314 (4.73), 375 (4.01), 473 (3.61) nm; IR (KBr) ν 3044, 2970, 2924, 2854, 1631, 1615, 1580, 1560, 1550, 1487, 1464, 1448, 1413, 1332, 1297, 1196, 1141, 1122, 912, 881, 858, 831, 743 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.56 (6H, s, CMe_2), 1.69 (2H, quintet, J=8, CH_2-2′), 2.16 (2H, td, J=8, 7, CH_2-3′), 2.71 (2H, t, J=8, CH_2-1′), 3.95 (3H, s, NMe), 4.97 (1H, dd, J=10, 2, H-5′a), 5.07 (1H, dd, J=17, 2, H-5′b), 5.54 (1H, d, J=10, H-2), 5.93 (1H, ddt, J=17, 10, 7, H-4′), 6.60 (1H, d, J=10, H-1), 7.42 (1H, t, J=8, H-10), 7.56 (1H, t, J=8, H-11), 7.69 (1H, s, H-13), 7.87 (1H, d, J=8, H-12), 7.99 (1H, d, J=8, H-9), 8.94 (1H, s, H-8), 14.83 (1H, s, OH); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 21.8 (C-1′), 27.1 (C($\underline{C}H_3$)$_2$), 28.3 (C-2′), 33.9 (C-3′), 44.3 (N$\underline{C}H_3$), 76.4 (C-3), 100.8 (C-14b), 105.8 (C-6a), 110.2 (C-5), 112.1 (C-13), 114.2 (C-5′), 122.1 (C-7a), 122.2 (C-1), 122.8 (C-2), 124.8 (C-10), 127.0 (C-12), 127.6 (C-8), 128.4 (C-8a), 128.8 (C-11), 129.6 (C-9), 136.4 (C-12a), 139.4 (C-4′), 141.9 (C-13a), 143.5 (C-14a), 160.2 (C-4a), 163.0 (C-6), 182.1 (C-7); ES-MS m/z 448 $[MNa]^+$, 426 $[MH]^+$. *Anal.* Calcd for $C_{28}H_{27}NO_3$: C,79.03; H, 6.40; N, 3.29. Found: C,78.94; H, 6.47; N, 3.22.

6-(5-Iodopentyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (13) Amorphous solid, UV λ (MeOH) (log ε) 206 (4.55), 241 (4.41), 276 (4.68), 298 (4.67), 307 (4.68), 364 (4.29), 443 (3.75) nm; IR (KBr) ν 3048, 2970, 2916, 2850, 1646, 1615, 1584, 1565, 1491, 1401, 1355, 1332, 1188, 1141, 1087, 889, 866, 803, 757, 737 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.55 (6H, s, CMe_2), 1.73 (2H, m, CH_2-3′), 1.95 (4H, m, CH_2-2′, CH_2-4′), 3.27 (2H, t, J=7, CH_2-5′), 3.89 (3H, s, NMe), 4.11 (2H, t, J=6, CH_2-1′), 5.53 (1H, d, J=10, H-2), 6.27 (1H, s, H-5), 6.57 (1H, d, J=10, H-1), 7.40 (1H, t, J=8, H-10), 7.53 (1H, t, J=8, H-11), 7.64 (1H, s, H-13), 7.85 (1H, d, J=8, H-12), 8.00 (1H, d, J=8, H-9), 8.93 (1H, s, H-8); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 6.9 (C-5′), 26.9 (C($\underline{C}H_3$)$_2$), 27.1, 28.0, 33.3 (3C, C-2′, C-3′, C-4′), 44.7 (N$\underline{C}H_3$), 68.7 (C-1′), 76.3 (C-3), 94.6 (C-5), 102.9 (C-14b), 109.6 (C-6a), 111.8 (C-13), 122.0 (C-1), 122.9 (C-2), 124.3 (C-10), 125.3 (C-7a), 126.6 (C-12), 128.1 (2C, C-8, C-11), 128.5 (C-8a), 129.5 (C-9), 135.6 (C-12a), 141.9 (C-13a), 147.7 (C-14a), 159.6 (C-4a), 162.5 (C-6), 177.8 (C-7); ES-MS m/z 592 $[MK]^+$, 576 $[MNa]^+$, 554 $[MH]^+$. *Anal.* Calcd for $C_{28}H_{28}INO_3$: C, 60.77; H, 5.10; N, 2.53; I, 22.93. Found: C, 60.87; H, 5.17; N, 2.49; I, 23.05.

8,8,11-Trimethyl-3,4,5,6,8,11-2*H*,18*H*-benzo[*i*]oxocino[2,3-*a*]-pyrano[2,3-*c*]acridin-18-one (17) Amorphous solid, UV λ (MeOH) (log ε) 208 (4.37), 241 (4.25), 282 (4.49), 312 (4.38), 354 (3.73), 447 (3.53) nm; IR (KBr) ν 3052, 2920, 2851, 1720, 1615, 1572, 1483, 1456, 1398, 1351, 1180, 1133, 1122, 1099, 1012, 744 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.56 (6H, s, CMe_2), 1.62 (6H, m, CH_2-3, CH_2-4, CH_2-5), 2.90 (2H, t, J=6, CH_2-6), 3.93 (3H, s, NMe), 4.33 (2H, m, CH_2-2), 5.60 (1H, d, J=10, H-9), 6.64 (1H, d, J=10, H-10), 7.41 (1H, t, J=8, H-15), 7.55 (1H, t, J=8, H-14), 7.68 (1H, s, H-12), 7.87 (1H, d, J=8, H-13), 8.02 (1H, d, J=8, H-16), 8.96 (1H, s, H-17); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 21.5 (C-6), 26.9 (C($\underline{C}H_3$)$_2$), 27.0, 29.0, 29.1 (3C, C-3, C-4, C-5), 44.9 (N$\underline{C}H_3$), 76.0 (C-8), 76.7 (C-2), 106.6 (C-10a), 111.9 (C-12), 112.5 (C-18a), 120.3 (C-6a), 122.4 (C-10), 124.2 (C-2), 124.5 (C-15), 125.0 (C-17a), 126.8 (C-13), 128.3 (2C, C-14, C17), 128.6 (C-16a), 129.7 (C-16), 135.9 (C-12a), 142.3 (C-11a), 145.6 (C-10b), 157.2 (C-6b), 158.8 (C-18b), 177.8 (C-18); ES-MS m/z 464 $[MK]^+$, 448 $[MNa]^+$. *Anal.* Calcd for $C_{28}H_{27}NO_3$: C, 79.03; H, 6.40; N, 3.29. Found: C, 79.10; H, 6.38; N, 3.37.

6,6′-(Pentane-1,5-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one) (9) Amorphous solid, UV λ (MeOH) (log ε) 206 (4.76), 242 (4.63), 276 (4.88), 297 (4.84), 307 (4.84), 362 (4.09), 441 (3.90) nm; IR (KBr) ν 3052, 2966, 2924, 2862, 1650, 1615, 1588, 1560, 1491, 1461, 1394, 1355, 1332, 1187, 1141, 1083, 759, 741 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.54 (12H, s, 2 CMe_2), 1.95 (2H, m, CH_2-3″), 2.14 (4H, m, CH_2-2″, CH_2-4″), 3.87 (6H, s, 2 NMe), 4.19 (4H, t, J=7, CH_2-1″, CH_2-5″), 5.51 (2H, d, J=10, H-2, H-2′), 6.31 (2H, s, H-5, H-5′), 6.56 (2H, d, J=10, H-1, H-1′), 7.38 (2H, t, J=8, H-10, H-10′), 7.51 (2H, t, J=8, H-11, H-11′), 7.61 (2H, s, H-13, H-13′), 7.84 (2H, d, J=8, H-12, H-12′), 7.97 (2H, d, J=8, H-9, H-9′), 8.93 (2H, s, H-8, H-8′); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 22.6 (C-3″), 26.9 (2 C($\underline{C}H_3$)$_2$), 28.7 (C-2″, C-4″), 44.7 (2 N$\underline{C}H_3$), 69.2 (C-1″, C-5″), 76.3 (C-3, C-3′), 94.9 (C-5, C-5′), 102.9 (C-14b, C-14b′), 109.8 (C-6a, C-6a′), 111.7 (C-13, C-13′), 122.0 (C-1, C-1′), 122.9 (C-2, C-2′), 124.3 (C-10, C-10′), 125.5 (C-7a, C-7a′), 126.7 (C-12, C-12′), 128.0 (C-11, C-11′), 128.2 (C-8, C-8′), 128.6 (C-12a, C-12a′), 129.6 (C-9, C-9′), 135.7 (C-8a, C-8a′), 142.1 (C-13a, C-13a′), 147.4 (C-14a, C-14a′), 159.8 (C-4a, C-4a′), 162.8 (C-6, C-6′), 177.8 (C-7, C-7′); ES-MS m/z 805 $[MNa]^+$, 783 $[MH]^+$. *Anal.* Calcd for $C_{51}H_{46}N_2O_6$: C, 78.24; H, 5.92; N, 3.58. Found C, 78.31; H, 6.01; N, 3.64.

Reaction of 6-Hydroxy-3,3-14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (6) with 1,6-Diiodohexane Alkylation of **6** (200 mg, 0.56 mmol) with 1,6-diiodohexane (286 μl, 1.75 mmol) was performed according to the procedure described for the condensation of **6** with 1,3-diiodopropane. Column chromatography over silica gel (eluent: CH_2Cl_2 then CH_2Cl_2/MeOH mixtures of increasing polarity) successively afforded 6-hydroxy-5-(6-iodohexyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (**22**) (33 mg, 10%), 6-(6-iodohexyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (**14**) (108 mg, 34%), and 6,6′-(hexane-1,6-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one) (**10**) (22 mg, 10%).

6-Hydroxy-5-(6-iodohexyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one (22) Amorphous solid, UV λ (MeOH) (log ε) 205 (4.43), 250 (4.37), 281 (4.68), 302 (4.73), 314 (4.74), 376 (4.02), 473 (3.62) nm; IR (KBr) ν 3054, 2970, 2924, 2851, 1634, 1615, 1580, 1558, 1490, 1463, 1413, 1331, 1300, 1192, 1141, 1126, 869, 749 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.42 (2H, m, CH_2), 1.56 (6H, s, CMe_2), 1.54—1.60 (4H, m, 2CH_2), 1.87 (2H, m, CH_2), 2.67 (2H, t, J=7, CH_2-1′), 3.21 (2H, t, J=7, CH_2-6′), 3.92 (3H, s, NMe), 5.54 (1H, d, J=10, H-2), 6.59 (1H, d, J=10, H-1), 7.40 (1H, t, J=8, H-10), 7.54 (1H, t, J=8, H-11), 7.65 (1H, s, H-13), 7.85 (1H, d, J=8, H-12), 7.96 (1H, d, J=8, H-9), 8.90 (1H, s, H-8), 14.82 (1H, s, OH); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 7.6 (CH_2-6′), 22.1 (CH_2-1′), 27.1 (C($\underline{C}H_3$)$_2$), 28.6, 28.7, 30.5, 33.7 (4C, CH_2-2′, CH_2-3′, CH_2-4′, CH_2-5′), 44.2 (N$\underline{C}H_3$), 76.4 (C-3), 100.8 (C-14b), 105.7 (C-6a), 110.3 (C-5), 112.1 (C-13), 122.1 (C-7a), 122.2 (C-1), 122.8 (C-2), 124.8 (C-10), 127.0 (C-12), 127.6 (C-8), 128.3 (C-8a), 128.8 (C-11), 129.6 (C-9), 136.4 (C-12a), 141.8 (C-13a), 143.4 (C-14a), 160.1 (C-4a), 163.0 (C-6), 182.1 (C-7); ES-MS m/z 606 $[MK]^+$, 590 $[MNa]^+$, 568 $[MH]^+$. *Anal.* Calcd for $C_{29}H_{30}INO_3$: C, 61.38; H, 5.33; N, 2.47; I, 22.36. Found: C, 61.41; H, 5.34; N, 2.51; I, 22.41.

6-(6-Iodohexyloxy)3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]-pyrano[3,2-*h*]acridin-7-one (14) Amorphous solid, UV λ (MeOH) (log ε) 204 (4.54), 242 (4.37), 276 (4.63), 298 (4.62), 307 (4.63), 363 (3.91); 443.2 (3.731) nm; IR (KBr) ν 3080, 3048, 2928, 2849, 1642, 1615, 1580, 1557, 1491, 1460, 1394, 1355, 1335, 1238, 1210, 1188, 1141, 1126, 1083, 1029, 990, 866, 831, 807, 745 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.54 (6H, s, CMe_2), 1.55 (2H, m, CH_2), 1.66 (2H, m, CH_2), 1.89 (2H, m, CH_2), 1.97 (2H, m, CH_2), 3.22 (2H, t, J=7, CH_2-6′), 3.85 (3H, s, NMe), 4.09 (2H, t, J=6, CH_2-1′), 5.50 (1H, d, J=10, H-2), 6.26 (1H, s, H-5), 6.55 (1H, d, J=10, H-1), 7.38 (1H, t, J=8, H-10), 7.50 (1H, t, J=8, H-11), 7.60 (1H, s, H-13), 7.82 (1H, d, J=8, H-12), 7.97 (1H, d, J=8, H-9), 8.92 (1H, s, H-8); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 7.2 (CH_2-6′), 26.8 (C($\underline{C}H_3$)$_2$), 24.9, 28.8, 30.2, 33.3 (4C, CH_2-2′, CH_2-3′, CH_2-4′, CH_2-5′), 44.6 (N$\underline{C}H_3$), 68.9 (CH_2-1′), 76.3 (C-3), 94.5 (C-5), 102.8 (C-14b), 109.6 (C-6a), 111.7 (C-13), 121.9 (C-1), 122.8 (C-2), 124.3 (C-10), 125.3 (C-7a), 126.6 (C-12), 128.0 (2C, C-8, C-11), 128.5 (C-8a), 129.4 (C-9), 135.6 (C-12a), 141.9 (C-13a), 147.4 (C-14a), 159.6 (C-4a), 162.6 (C-6), 177.7 (C-7); ES-MS m/z 606 $[MK]^+$, 590 $[MNa]^+$, 568 $[MH]^+$. *Anal.* Calcd for $C_{29}H_{30}INO_3$: C, 61.38; H, 5.33; N, 2.47; I, 22.36. Found: C, 61.46; H, 5.29; N, 2.45; I, 22.44.

6,6′-(Hexane-1,6-diylbisoxy)bis(3,3,14-trimethyl-3,14-dihydro-7*H*-benzo[*b*]pyrano[3,2-*h*]acridin-7-one) (10) Amorphous solid, UV λ (MeOH) (log ε) 205 (4.66), 241 (4.52), 275 (4.74), 297 (4.67), 357 (3.98), 434 (3.77) nm; IR (KBr) ν 3052, 2920, 2854, 1646, 1615, 1588, 1557, 1487, 1460, 1394, 1355, 1332, 1184, 1141, 1122, 1081, 741 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 1.54 (12H, s, 2 CMe_2), 1.75 (4H, m, CH_2-3″, CH_2-4″), 2.06 (4H, m, CH_2-2″, CH_2-5″), 3.90 (6H, s, 2 NMe), 4.16 (4H, t, J=6, CH_2-1″, CH_2-6″), 5.51 (2H, d, J=10, H-2, H-2′), 6.31 (2H, s, H-5, H-5′), 6.57 (2H, d, J=10, H-1, H-1′), 7.40 (2H, t, J=8, H-10, H-10′), 7.53 (2H, t, J=8, H-11, H-11′), 7.64 (2H, s, H-13, H-13′), 7.86 (2H, d, J=8, H-12, H-12′), 8.00 (2H, d, J=8, H-9, H-9′), 8.95 (2H, s, H-8, H-8′); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 25.8 (C-3″, C-4″), 26.9 (2 C($\underline{C}H_3$)$_2$), 28.9 (C-2″, C-5″), 44.8 (2 N$\underline{C}H_3$), 69.2 (C-1″, C-6″), 76.3 (C-3, C-3′), 94.7 (C-5, C-5′), 102.8 (C-14b, C-14b′), 109.7 (C-6a, C-6a′), 111.7 (C-13, C-13′), 122.1 (C-1, C-1′), 122.9 (C-2, C-2′), 124.3 (C-10, C-10′), 125.4 (C-7a, C-7a′), 126.7 (C-12, C-12′), 128.1 (C-11, C-11′), 128.2 (C-8, C-8′), 128.6 (C-12a, C-12a′), 129.6 (C-9, C-9′), 135.7 (C-8a, C8-a′), 142.0 (C-13a, C-13a′), 147.5 (C-14a, C-14a′), 159.8 (C-4a, C-4a′), 162.8 (C-6, C-6′), 177.8 (C-7, C-7′); ES-MS m/z 835

$[MK]^+$, 819 $[MNa]^+$. *Anal.* Calcd for $C_{52}H_{48}N_2O_6$: C, 78.37; H, 6.07; N, 3.52. Found: C, 78.45; H, 6.12; N, 3.52.

Pharmacology. Cytotoxicity Murine leukemia L1210 cells from the American Type Culture Collection (Rockville Pike, MD, U.S.A.) were grown in RPMI medium 1640 supplemented with 10% fetal calf serum, 2 mM L-glutamine, penicillin 100 U/ml, streptomycin 100 μg/ml and 10 mM HEPES buffer (pH 7.4). The cytotoxicity was measured using the microculture tetrazolium assay essentially as described.[29] Cells were exposed for 48 h to nine graded concentrations in triplicate of the test drug. Results are expressed as IC_{50} values (mean, $n=3$), which are defined as the drug concentration inhibiting the absorbance by 50% with respect to that of untreated cells.

Cell Cycle Analysis For the cell cycle analysis, L1210 cells (5×10^5 cells/ml) were incubated for 21 h with various concentrations of the drug. Cells were then fixed by 70% ethanol (v/v), washed, and incubated in PBS containing 100 μg/ml RNAse and 50 μg/ml propidium iodide for 30 min at 20 °C. For each sample, 10000 cells were analyzed on an XLMCL flow cytometer (Beckman Coulter, France). Results are expressed as % of cells arrested in the given phases of the cell cycle.

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