Bioorganic & Medicinal Chemistry 18 (2010) 242–248

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry

journal homepage: www.elsevier.com/locate/bmc

Multivalent catanionic GalCer analogs derived from first generation dendrimeric phosphonic acids

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ARTICLE INFO

Article history:
Received 18 September 2009
Revised 27 October 2009
Accepted 29 October 2009
Available online 3 November 2009

Keywords:
Dendrimer
Galcer
Galactosylceramide

ABSTRACT

The synthesis and characterization of a new series of catanionic multivalent analogs of GalCer is described. These systems are based on phosphonic acid terminated dendrimers and *N*-hexadecylamino lactitol moieties. Despite important structural differences that affect the dendrimers' outer-shell, these supramolecular assemblies showed a fairly comparable anti-HIV-1 activity. All compounds have submicromolar IC_{50} in a cell-based HIV-infection model but also a high general cytotoxicity.

1. Introduction

Dendrimers are receiving a constantly increasing attention since the first synthesis was published by Vögtle and his co-workers.[1] Their globular and well-defined structure and the relative ease of discrete functionalization of their architecture make these macromolecules attractive scaffolds for biomedical applications.[2] Indeed, water-soluble dendrimers are efficient carriers of active substances that can be entrapped within the dendrimer's architecture[3] or formulated with the latter.[4] Another option is to graft covalently a drug or a pro-drug on the surface of a dendrimer,[5] either with a complete capping of the surface, or in a statistical manner,[6] leaving free sites to graft other molecules of interest like targeting moieties and/or fluorescent tags.

In the specific field of dendrimers designed for antiviral purposes,[7] these strategies based on covalent grafting or cargo loading have also been developed with a special emphasis on multivalency.[8,9] For instance, poly(propyleneimine) (PPI) tuftsin-dendrimer conjugates, a natural phagocytosis stimulating peptide that binds to macrophages, have been loaded with efavirenz.[10] The same authors have loaded mannose capped PPI dendrimers with antiretroviral drug lamivudine and observed a significant increase of its activity, cellular uptake and a reduced cytotoxicity.[11] Other strategies include the use of dendrimers to transfect suitable silencing RNA to infected lymphocytes and reduce HIV replication.[12]

The highly glycosylated glycoprotein gp120 present on the surface HIV-1 is also a key target to block the virus infection. Its recognition by CD4(+) and the subsequent formation of a ternary complex involving gp120, CD4 and co-receptors (namely CCR5 and CXCR4)[13] is a pivotal step of the infection. In this regard, Schengrund et al. have reported on randomly sulfonated galactose-terminated PPI dendrimers[14,15] as efficient in vitro binding antagonists for HIV-1. Recently, dendrimeric oligomannose glycans have been designed to mimic a mannose cluster identified as the epitope recognized by the HIV-1-neutralizing monoclonal antibody 2G12. These dendrimeric oligomannose glycans were found to strongly inhibit the binding of gp120 to 2G12.[16] The virus can also infect CD4(−) cells through gp120 binding to galactosylceramide (GalCer).[17] It has been shown that GalCer is directly interacting with gp41,[18] and that gp120 is also expressed at the surface of infected cells.[19] Consequently, the gp120/GalCer interactions have become a relevant target in view of blocking both CD4(+)-mediated and alternative CD4(−)-mediated infection pathways.[20,21] In this respect, anionic polymers[22] and dendrimers[23] have been used to inhibit HIV-1 infection by preventing virus-cell fusion through ionic interactions with the V3 loop of gp120 and led to barrier gels for topical use that are currently under clinical trials,[24] like Vivagel, a gel based on poly-L-lysine dendrimers with naphtalene sulfonic

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doi:10.1016/j.bmc.2009.10.058

terminations.[25] These gels are based on polyanionic compounds, and to the best of our knowledge, none of them was designed with specific targeting moieties able to disrupt the HIV-1-CD4 or HIV-1-GalCer interactions.

In the past, we have developed a simple approach to design multivalent GalCer analogs[4,26] based on catanionic systems,[27–30] that proved to be good HIV-1 inhibitors. The influence of the core functionality of the PPH (poly(phosphorhydrazone)) dendrimeric structure[31] was clearly identified, and the bioactivities were found to be core-dependent but not generation dependent. Noteworthy, the influence of the outer-shell had not been studied to date. In this respect, we present here a collection of new multivalent catanionic systems based on various first generation phosphonic acid terminated dendrimers and *N*-hexadecylamino-lactitol moieties in order to evaluate the direct influence of the vicinity of the phosphonic acid on the HIV-1 inhibitory properties of both the corresponding mono sodium salts and the related catanionic assemblies.

2. Results and discussion

2.1. Chemistry

The general synthetic strategy is based on the conversion of dimethylphosphonate terminated dendrimers **1a-G1** to **6a-G1** (G stands for generation) to the corresponding poly-phosphonic acids **1b-G1** to **6b-G1** by means of a silylation-methanolysis procedure (Fig. 1).[32] The reaction proceeds cleanly in dilute acetonitrile solutions (1–10 mmol L^{-1}) in 24 h at room temperature, using 2.5–3 equiv of bromotrimethylsilane per phosphonate.[33] In many cases, attempts to shorten the reaction time by a temperature elevation resulted in partial degradation of the dendrimeric structure. It is worth noting that the reaction can also be run in dichloromethane without major change. The acidic dendrimers were then converted to the corresponding mono sodium salts **1c-G1** to **6c-G1** (Fig. 1) by addition of the stoichiometric amount of sodium hydroxide to a suspension of the acidic dendrimers in water. Alternatively, the catanionic assemblies **1d-G1** to **6d-G1** (Fig. 2) were obtained quantitatively by adding a stoechiometric amount of *N*-hexadecylamino-lactitol to the dendrimers suspended in water or in a water–propanol mixture. All compounds were fully characterized by multinucleus NMR. As previously reported, mass spectrometry is not compatible with the PPH (poly-phosphorhydrazone) dendrimeric structures[34] used in this study, nevertheless this technique was used for key intermediate compounds.

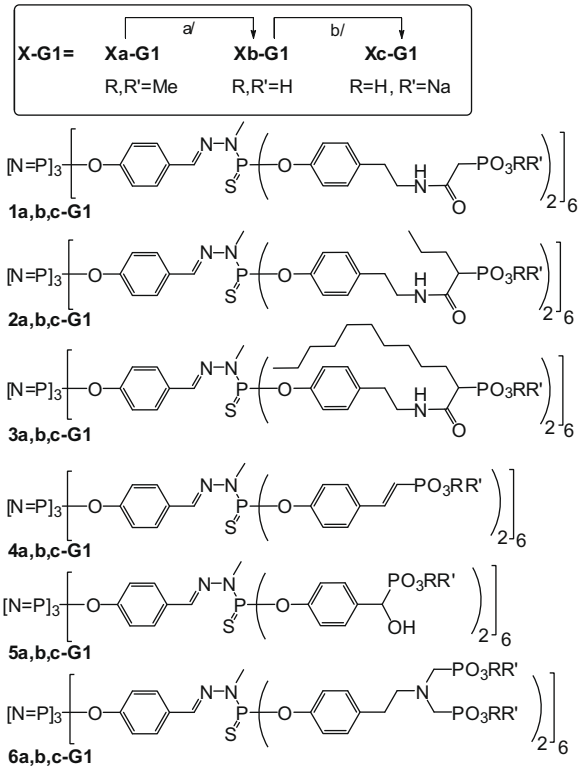


Figure 1. Collection of phosphonate-terminated dendrimers used as multivalent catanionic GalCer precursors ((a) $BrSiMe_3$, MeOH; (b) HONa).

A first series of PPH dendrimers **1-G1** to **3-G1** bearing phosphonic acid moieties and lateral alkyl chains was prepared according to a phenol grafting procedure involving the use of tyramine-based dimethyl-phosphonoacetate with a pendant alkyl chain of variable length on the activated methylene position.[35] The dendrimers **1a-G1** to **3a-G1** were converted to the corresponding phosphonic acids monosodium salts **1c-G1** to **3c-G1**.

The dendrimeric catanionic systems were obtained by mixture of the acidic terminated dendrimers with 12 equiv of *N*-hexadecylamino-lactitol in water. The reaction mixture evolutes from a heterogeneous mixture, in which the phosphonic acid terminated dendrimers are suspended, to a turbid and then clear solution. Interestingly, the effect of the alkyl chain on the formation of the catanionic assemblies was not linear. Indeed, **1d-G1** and **3d-G1** were obtained in 24 h, whereas **2d-G1** was obtained in three days. In this respect, we have recently shown[35] that the interactions of the C3 alkyl chains of **2c-G1** with the interior of the dendrimer structure were weaker than the interactions of the C10 alkyl chains of **3c-G1** with the same structure. This phenomenon can be explained by the localization of the alkyl chains of **3b-G1** which can be assumed to be more tightly buried within the interior of the structure. On the contrary, the C3 alkyl chains of **2b-G1** are less pre-organized within the dendrimeric structure and have more degrees of freedom. Consequently, the formation of the assembly **2d-G1** may be entropically more demanding. As expected for these phosphorus-containing structures, all reactions were easily monitored by ^{31}P NMR (Table 1), for instance the conversion of the parent phosphonate moieties to the phosphonic acids is accompanied on the ^{31}P NMR spectra by a shielding of the corresponding singlet from 28.2 ppm to 23.2 ppm in the case of **2-G1**. Upon addition of 12 equiv of sodium hydroxide this singlet is shielded to 17.4 ppm for **2c-G1**, while the formation of **2d-G1** is traduced by the appearance of a singlet at 16.8 ppm. Remarkably, the other regions of the spectrum, namely the singlets located in the 8–9 ppm and 62–64 ppm regions and attributed to the cyclotriphosphazene core and the PS atoms of the divergent points, respectively, are not affected by the whole procedure.

A new vinyl-dimethylphosphonate terminated dendrimer **4a-G1** was also prepared, and successfully converted into the corresponding vinyl-phosphonic acid capped dendrimer **4b-G1**. The first step involves a Horner–Wadsworth–Emmons reaction between the aldehyde terminated PPH dendrimer **G1** and tetramethyl-methylene-*gem*-diphosphonate (Scheme 1) with sodium hydride as a base. The reaction proceeds cleanly at room temperature, and the removal of side product sodium dimethylphosphate was performed by column chromatography. Again, the reaction was followed by NMR, allowing us to observe the total disappearance of the signals corresponding to the aldehyde proton of **G1** ($\delta(^1H)$ = 9.97 ppm, $\delta(^{13}C)$ = 189.9 ppm), and the appearance of two sets of doublets on the ^1H NMR spectrum at 6.10 ppm and 7.35 ppm, with a typical $^3J_{HHtrans}$ = 17.4 Hz, corresponding, respectively to the C_6H_4CH and CH–P proton of the vinyl group. The next step was again followed by ^{31}P NMR, which shows the total disappearance of the singlet located at 21.9 ppm attributed to the PO_3Me_2 moieties of **4a-G1** and the appearance of a singlet at 13.6 ppm corresponding to the PO_3H_2 moieties of **4b-G1**. The

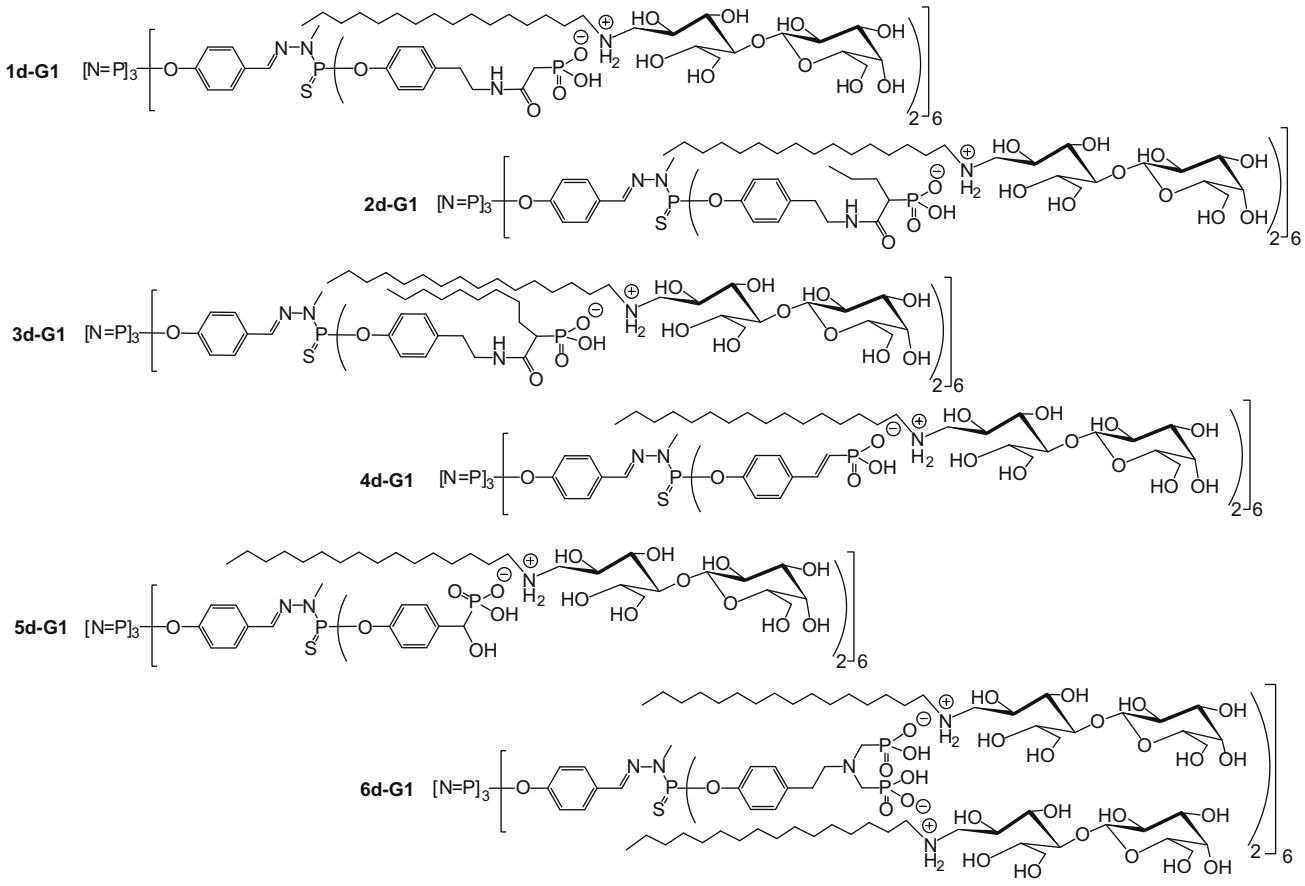
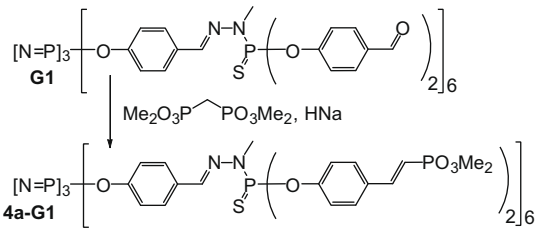


Figure 2. Collection of multivalent catanionic systems based on phosphonate-terminated dendrimers and hexadecylamino lactitol.

Table 1
^{31}P NMR chemical shifts for the phosphorus atoms of compounds **1n-G1** to **6n-G1** (n = **a** to **d**)

Entry	Compound	δ ^{31}P ($CDCl_3$, n = **a**)			δ ^{31}P (CD_3OD, n = **b**)			δ ^{31}P (D_2O/CD_3CN, n = **c**)			δ ^{31}P (D_2O/CD_3CN, n = **d**)		
		[P=N]$_3$	PO_3Me_2	P(S)	[P=N]$_3$	PO_3H_2	P(S)	[P=N]$_3$	PO_3HNa	P(S)	[P=N]$_3$	PO_3HL	P(S)
1	**1n-G1**	8.4	25.4	62.8	9.3	19.5	62.7	9.7	13.8	64.0	9.2	13.4	63.6
2	**2n-G1**	8.4	28.2	62.9	9.3	23.2	62.8	9.9	17.4	64.0	9.4	16.8	63.7
3	**3n-G1**	8.4	28.1	63.0	9.3	22.4	62.7	9.0	17.8	64.0	9.2	17.3	62.8
4	**4n-G1**	8.2	21.9	62.9	8.2	13.6	62.2	9.8	12.5	63.6	9.1	11.0	62.8
5	**5n-G1**	8.9	23.9	62.4	9.1	20.1	62.8	9.0	15.9	64.8	8.6	15.9	63.5
6	**6n-G1**	8.3	26.9	63.1	9.5	10.4	66.3	9.6	6.8	64.2	8.6	7.2	63.0



Scheme 1. Synthesis of dendrimer **4a-G1**.

monosodium salt **4c-G1** and the catanionic assembly **4d-G1** were easily obtained as described above, and the chemical shifts observed on the ^{31}P spectra, along with ^{1}H and ^{13}C NMR analysis were consistent with the expected structures. In particular, the phosphorus atoms of the phosphonate groups were found to resonate at 12.5 and 11.0 ppm, respectively on the ^{31}P NMR spectra. The series of dendrimers **5a-G1** to **5c-G1** and **6a-G1** to **6c-G1** having α-hydroxyphosphonate and bismethylene-phosphonate end-groups, respectively were synthesized[32] according to a similar strategy. Similarly to previous series, the ^{31}P NMR was intensively used to follow all reactions, and the expected catanionic dendrimers **5d-G1** and **6d-G1** presenting, respectively 12 and 24 GalCer analogs on their outer-shell were readily obtained and fully characterized by multinucleus NMR.

2.2. HIV-1 inhibitory activity

The **Xc-G1** (X = 1–6) series of mono sodium salt compounds were assayed in vitro on a CEM-SS cell-line to assess their inhibitory effect against HIV-1 (Table 2, entry 1–6).[35–38] Interestingly, none of these compounds was found to be toxic in the whole range of concentrations (1×10^{-7} to 1×10^{-4} mol L^{-1}). Consequently, the measured IC_{50} can be correlated to an effective antiviral activity. Among this series, the IC_{50} values were found to slightly depend on the chemical nature of phosphonate's vicinity. The influence of the alkyl chain (Table 2, entries 1–3) has already been

Table 2
Classical HIV-1 inhibition and cytotoxicity results for compounds **1c-G1** to **6c-G1**, and **1d-G1** to **6d-G1**

Entry	Compound	HIV-1 inhibition IC_{50}, μM[a]	Cytotoxicity CC_{50}, μM[a]	T.I.[b]
1	**1c-G1**	25[35]	>100	>4
2	**2c-G1**	1.5[35]	>100	>67
3	**3c-G1**	16[35]	>100	>6
4	**4c-G1**	0.65	>100	>153
5	**5c-G1**	50	>100	>2
6	**6c-G1**	20	>100	>5
7	**1d-G1**	0.40	1.9	4.8
8	**2d-G1**	0.33	1.5	4.5
9	**3d-G1**	0.31	6.1	19.7
10	**4d-G1**	0.25	1.6	6.4
11	**5d-G1**	0.21	1.2	4
12	**6d-G1**	0.16	1.1	6.9

[a] Values are means of two or three experiments, standard deviation 10%.
[b] In vitro therapeutic index (CC_{50}/IC_{50}).

evoked in a previous paper, and was assumed to be linked to its propensity to interact with the lipophilic region of the V3 loop of gp120.[35] Compounds **5c-G1** and **6c-G1** (Table 2, entry 5 and 6) show a relatively low HIV-1 inhibitory activity, in the range of that of **1c-G1** and **3c-G1** (Table 2, entry 1 and 3), whereas the vinylphosphonate terminated dendrimer (Table 2, entry 4) exhibit a good inhibitory activity (650 nmol L^{-1}) which is in the range of activity of two candidate polyanion compounds in clinical trials,[39] PRO 2000 and dextrin sulphate (DxS). The Therapeutic Index of **4c-G1** measured by default at 153 is one order of magnitude lower than the target value of 1000,[40] whereas all other compounds, for which the therapeutic indices (TIs) are too low, might induce mucosal irritation or inflammation that can be responsible for an increased HIV transmission.[41]

The **Xd-G1** (X = 1–6) series of catanionic multivalent GalCer analog compounds were assayed according to the same procedure (Table 2, entries 7–12). All compounds exhibit a relatively good anti-HIV-1 activity between 0.16 μM (Table 2, entry 12) and 0.40 μM (Table 2, entry 7), but their TIs is dramatically affected by CC_{50} values that are less than tenfold the value of IC_{50}, except in case of **3d-G1** (Table 2, entry 9). The fact that the latter present a lower toxicity than its concurrent multivalent analogs is not fully understood. One possible explanation could reside in the possible lack of stability of the supramolecular assemblies in cell cultures. As a result, the system could eventually disassemble into dendrimeric phosphonates for which no cytotoxicity has been observed (Table 2, entries 1–6), and free *N*-hexadecylaminolactitol that could be responsible for the relatively high CC_{50}. Assuming that a putative complete disassembly of the catanionic systems would lead to 12 free *N*-hexadecylaminolactitol, the relative CC_{50} of the dissembled **Xd-G1** series should then be corrected by a factor of 12-fold, leading to CC_{50} that are in the range of *N*-hexadecylamino lactitol which is known to be cytotoxic on CEM-SS cells in the 10^{-4}–10^{-5} mol L^{-1} range due to its detergent properties.[4] Accordingly, the supramolecular assembly **3d-G1** can be assumed to be more stable than the other thanks to the presence of a C10 alkyl chain close to the phosphonic acid, which could consolidate the lipophilic interaction between this C10 alkyl chain and the alkyl chain of the *N*-hexadecylaminolactitol.

3. Conclusions

We have described the preparation of a series of dendrimeric GalCer analogs based on catanionic assemblies of *N*-hexadecylaminolactitol moieties and phosphonic acid dendrimers. These compounds, along with their mono sodium salts and dendrimeric precursors have been extensively studied by multinucleus NMR. In particular, the ^{31}P NMR has proven to be an efficient tool to monitor all chemical transformations, and the collected chemical shifts of the terminal phosphonate (or phosphonic acid) moieties gathered in Table 1 present a coherent set of chemical shifts in agreement with previous works.[42] The in vitro HIV-1 inhibitory properties on CEM-SS cells of the monosodium salt series are in agreement with previous observations for polyanionic dendrimers or linear polymers,[24] but all compounds suffer from low TIs values that prevent them from being so-called promising candidates. Nevertheless, our results show unambiguously that the local environment of the negative charges, that is the chemical nature of the phosphonate neighboring groups, strongly influences the inhibitory properties. On the contrary, this environment has a lower impact on the biological activity of catanionic multivalent analogs. Actually, the GalCer analog series show good HIV-1 inhibitory properties, but quite low TIs were measured for these compounds, due to relatively high CC_{50} values, probably related to a lack of stability of the assemblies in vitro. Further experiments are currently under study to assess the fact that the length of an alkyl chain located close to the phosphonic acid end-groups could increase the in vitro stability of the supramolecular assembly. In this respect, multivalent GalCer systems based on supramolecular assemblies appear as a valuable strategy to develop original anti-HIV-1 candidates, given that their in vitro stability can be significantly increased.

4. Methods

4.1. Chemistry

4.1.1. General

All manipulations were carried out using standard high-vacuum and dry-argon techniques. Chemicals were used as received and solvents were dried and distilled by routine procedures.[43] ^{1}H, ^{13}C, ^{19}F and ^{31}P NMR spectra were recorded at 25 °C with Bruker AC 200, AV 300, DPX 300, AV 400 or AV500 spectrometers. The following abbreviations were used in reporting spectra: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, dd = doublet of doublets. References for NMR chemical shifts are 85% H_3PO_4 for ^{31}P NMR and $SiMe_4$ for ^{1}H and ^{13}C NMR. The attribution of ^{13}C NMR signals was done using *J*mod, 2D ^{1}H–^{13}C HSQC, ^{1}H–^{13}C HMBC and ^{1}H–^{31}P HMQC, Broad Band or CW ^{31}P decoupling experiments when necessary. Mass spectrometry was recorded on a Finnigan-mat TSQ 7000. The numbering schemes used for NMR are depicted in Figure 3. PPH dendrimer precursors[44,45] and *N*-hexadecylaminolactitol[46] were prepared according to published procedure.

4.1.2. Synthesis of phosphonate capped dendrimers Xa-G1 (X = 1–6)

Dendrimers **1a-G1** to **3a-G1**,[35] **5a-G1** and **6a-G1** were prepared according to published procedure.[32] Dendrimer **4a-G1** was prepared as follows: to a suspension of NaH (141 mg, 5.840 mmol) in THF (10 mL) was added dropwise tetramethyl-methylene-*gem*-diphosphonate. Then, this solution was dropped into a solution of **G1** (1383 mg, 5.840 mmol) in THF (5 mL) at 0 °C. The mixture was stirred at rt overnight and was evaporated to dryness. The white residue was diluted in water (150 mL) and extracted with chloroform (3 × 50 mL). The organic phase was washed with brine (50 mL), separated, dried over Na_2SO_4, filtered and concentrated to dryness under reduced pressure. The residue was purified by silica gel flash chromatography (CH_2Cl_2/MeOH, 95:5, R_f = 0.26) to obtain **4a-G1** as a white powder (779 mg, 54%). ^{31}P {^{1}H} NMR ($CDCl_3$, 75.5 MHz): δ = 8.2 (s, N_3P_3), 21.9 (s, PO_3Me_2), 61.9 (s, P═S) ppm; ^{1}H NMR ($CDCl_3$, 300.1 MHz): δ = 3.28 (d, $^{3}J_{PH}$ = 9.6 Hz, 18H, NCH_3), 3.74 (d, $^{3}J_{PH}$ = 10.8 Hz, 72H, OCH_3), 6.10 (dd, $^{2}J_{PH} = {}^{3}J_{HH}$ = 17.4 Hz, 12H, PCH), 7.01 (d, $^{3}J_{HH}$ = 6.3 Hz, 12H, C_0^2H),

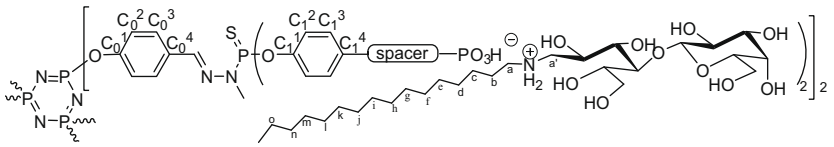


Figure 3. Numbering scheme for catanionic dendrimers.

7.17 (d, $^3J_{HH}$ = 6.9 Hz, 24H, C_1^2H), 7.35 (t, $^3J_{HH}$ = 20.7 Hz, 12H, ArC*H*), 7.39 (d, $^3J_{HH}$ = 6.9 Hz, 24H, C_1^3H), 7.61 (br d, $^3J_{HH}$ = 8.1 Hz, 18H, C_0^3H, CH=N) ppm; ^{13}C {^{1}H} NMR ($CDCl_3$, 75.5 MHz): δ = 33.0 (d, $^2J_{PC}$ = 12.1 Hz, NCH_3), 52.5 (d, $^2J_{PC}$ = 5.3 Hz, OCH_3), 112.8 (d, $^1J_{PC}$ = 192.5 Hz, PCH), 121.4 (s, C_0^2), 121.8 (d, $^3J_{PC}$ = 4.5 Hz, C_1^2), 128.3 (s, C_0^3), 129.1 (s, C_1^3), 132.0 (s, C_1^4), 132.3 (s, C_0^4), 139.0 (d, $^3J_{PC}$ = 15.1 Hz, N=CH), 148.1 (d, $^2J_{PC}$ = 6.8 Hz, ArC*H*), 151.4 (s, C_0^1), 151.9 (d, $^2J_{PC}$ = 6.8 Hz, C_1^1) ppm.

4.1.3. Synthesis of phosphonic acid capped dendrimers Xb-G1 (X = 1–6)

Dendrimers **1a-G1** to **3a-G1**,[35] **5a-G1** and **6a-G1** were prepared according to published procedure.[32] Dendrimer **4b-G1** was prepared as follows: to a vigorously stirred solution of **4a-G1** (200 mg, 0.048 mmol) in dry acetonitrile (10 mL) bromotrimethylsilane (192 μL, 1.440 mmol) was added at 0 °C. The mixture was stirred for 12 h at rt, and then evaporated to dryness under reduced pressure. The crude residue was washed twice with methanol (15 mL) for 1 h at rt and evaporated to dryness under reduced pressure. The resulting white solid was washed with water, methanol and Et_2O to afford **4b-G1** as a white solid (57 mg, 31%). ^{31}P {^{1}H} NMR (DMSO-d_6, 121.5 MHz): δ = 8.2 (s, N_3P_3), 13.6 (s, PO_3Me_2), 62.2 (s, P=S) ppm; ^{1}H NMR (DMSO-d_6, 300.1 MHz): δ = 3.29 (d, $^3J_{PH}$ = 10.8 Hz, 18H, NCH_3), 6.41 (dd, $^2J_{PH}$ = $^3J_{HH}$ = 18.2 Hz, 12H, PCH), 7.01–7.25 (m, 48H, C_0^2H, C_1^2H and ArC*H*), 7.56 (d, $^3J_{HH}$ = 8.4 Hz, 24H, C_1^3H), 7.64 (d, $^3J_{HH}$ = 8.4 Hz, 12H, C_0^3H), 7.86 (br s, 6H, CH=N) ppm; ^{13}C {^{1}H} NMR (DMSO-d_6, 75.5 MHz): δ = 33.3 (d, $^2J_{PC}$ = 11.9 Hz, NCH_3), 121.3 (d, $^1J_{PC}$ = 181.6 Hz, PCH), 121.4 (s, C_0^2), 121.7 (br s, C_1^2), 128.8 (s, C_0^3), 129.4 (s, C_1^3), 132.4 (s, C_0^4), 133.3 (d, $^3J_{PC}$ = 22.4 Hz, C_1^4), 141.0 (br s, N=CH), 142.1 (br s, ArC*H*), 151.0 (s, C_1^1), 151.1 (s, C_0^1) ppm.

4.1.4. Synthesis of sodium phosphonate capped dendrimers Xc-G1 (X = 1–6)

Dendrimers **1a-G1** to **3a-G1**,[35] **5a-G1** and **6a-G1** were prepared according to published procedure.[32] Dendrimer **4c-G1** was prepared as follows: the sodium monosalt form was obtained by adding aqueous sodium hydroxide (0.1023 N, 3.09 mL) to a suspension of **4b-G1** (100 mg, 0.026 mmol) in water (5 mL) at 0 °C. The solution was filtered on micropore (1.2 μm) and lyophilized to give **4c-G1** as a white powder (96 mg, 91%). ^{31}P {^{1}H} NMR (D_2O/CD_3CN, 3:1, 121.5 MHz): δ = 8.2 (s, N_3P_3), 12.5 (s, PO_3Me_2), 63.6 (s, P=S) ppm; ^{1}H NMR (D_2O/CD_3CN, 3:1, 300.1 MHz): δ = 3.28 (d, $^3J_{PH}$ = 9.0 Hz, 18H, NCH_3), 6.36 (dd, $^2J_{PH}$ = $^3J_{HH}$ = 15.0 Hz, 12H, PCH), 7.00–7.08 (m, 48H, C_0^2H, C_1^2H and ArC*H*), 7.45 (d, $^3J_{HH}$ = 6.0 Hz, 24H, C_1^3H), 7.61 (br s, 12H, C_0^3H), 7.82 (br s, 6H, CH=N) ppm; ^{13}C {^{1}H} NMR (D_2O/CD_3CN, 3:1, 125.8 MHz): δ = 32.8 (br s, NCH_3), 113.9 (br s, PCH), 121.4 (br s, C_1^2), 122.5 (br s, C_0^2), 124.0 (s, C_0^3), 128.6 (s, C_1^3), 132.2 (s, C_0^4), 134.1 (s, C_1^4), 139.8 (br s, ArCH and CH=N), 151.4 (br s, C_0^1 and C_1^1) ppm.

4.1.5. Synthesis of catanionic dendrimers Xd-G1 (X = 1–6)

1d-G1: Dendrimer **1b-G1** (50 mg, 0.011 mmol) was added to a solution of *N*-hexadecylaminolactitol (75 mg, 0.132 mmol) in distilled water (10 mL). After 3 days stirring at rt, the colorless homogeneous solution was freeze-dried to afford **1d-G1** as a white powder (124 mg, 100%). ^{31}P {^{1}H} NMR (D_2O/CD_3CN, 3:1, 121.5 MHz): δ = 9.2 (s, N_3P_3), 13.8 (s, PO_3Me_2), 64.0 (s, P=S) ppm; ^{1}H NMR (D_2O/CD_3COCD_3, 1:3, 500.1 MHz): δ = 0.77 (br s, 36H, CH_3), 1.12 (br s, 312 H, $C_{c\text{-}o}H_2$), 1.60 (br s, 24H, C_bH_2), 2.56–2.68 (m, 48H, ArCH_2 and PCH_2), 2.93 (br s, 48H, C_aH and $C_{a'}H$), 3.17–3.28 (m, 42H, NHCH_2 and NCH_3), 3.41–3.99 (m, 156H, CHOH, CH_2OH and CH_{anom}), 6.83 (br s, 12H, C_0^2H), 7.02 (br s, 24H, C_1^2H), 7.09 (br s, 24H, C_1^3H), 7.59 (br s, 12H, C_0^3H), 7.69 (br s, 6H, CH=N) ppm; ^{13}C NMR (D_2O/CD_3CN, 3:1, 75.5 MHz): δ = 14.1 (s, CH_3), 22.9 (s, C_o), 26.2 (s, C_b), 26.9 (s, C_c), 29.8 (s, C_d), 30.2 (br s, $C_{e\text{-}m}$), 32.2 (s, C_n), 32.9 (d, $^2J_{PC}$ = 19.5 Hz, CH_3N), 34.3 (s, $ArCH_2$), 38.3 (d, $^1J_{PC}$ = 197.3 Hz, PCH), 41.2 (s, $NHCH_2$), 42.3 (s, C_a), 43.5 (s, $C_{a'}$), 60.4 (s, CH_2OH), 61.2 (s, CH_2OH), 68.7 (s, CHOH), 70.3 (s, CHOH), 71.4 (s, CHOH), 71.5 (s, CHOH), 72.8 (s, CHOH), 75.5 (s, CHOH), 78.7 (s, CHO), 78.9 (s, CHO), 103.1 (s, C_{anom}), 121.3 (s, C_1^2), 121.6 (s, C_0^2), 128.6 (s, C_0^3), 130.3 (s, C_1^3), 132.7 (s, C_0^4), 137.2 (s, C_1^4), 140.0 (br s, CH=N), 148.9 (d, $^2J_{PC}$ = 7.6 Hz, C_1^1), 151.2 (s, C_0^1), 171.1 (s, CONH) ppm.

2d-G1: Dendrimer **2b-G1** (50 mg, 0.010 mmol) was added to a solution of *N*-hexadecylaminolactitol (68 mg, 0.120 mmol) in distilled water (10 mL). After 3 days stirring at rt, the colorless homogeneous solution was freeze-dried to afford **2d-G1** as a white powder (118 mg, 100%). ^{31}P {^{1}H} NMR (D_2O/CD_3CN, 3:1, 121.5 MHz): δ = 9.4 (s, N_3P_3), 16.8 (s, PO_3Me_2), 63.7 (s, P=S) ppm; ^{1}H NMR ($D_2O/CD_3(CO)CD_3$, 1:3, 500.1 MHz): δ = 0.90 (t, $^3J_{HH}$ = 6.5 Hz, 36H, CH_3), 0.94 (t, $^3J_{HH}$ = 6.5 Hz, 36H, CH_3), 1.24–1.44 (br s, 360H, CH_3CH_2 and $C_{c\text{-}o}H_2$), 1.53–1.88 (m, 48H, $CHCH_2$ and C_bH), 2.68–2.91 (m, 36H, $ArCH_2$ and CHP), 3.06–3.17 (m, 48H, C_aH_2 and $C_{a'}H_2$), 3.18–3.29 (m, 18H, NCH_3), 3.32–3.48 (m, 48H, $NHCH_2$), 3.58–4.08 (m, 120H, CHOH and CH_2OH), 4.31 (br s, 12H, CH_{anom}), 6.91 (d, $^3J_{HH}$ = 8.0 Hz, 12H, C_0^2H), 7.11 (d, $^3J_{HH}$ = 8.0 Hz, 24H, C_1^2H), 7.17 (d, $^3J_{HH}$ = 8.5 Hz, 24H, C_1^3H), 7.67 (d, $^3J_{HH}$ = 7.0 Hz, 12H, C_0^3H), 7.77 (br s, 6H, CH=N) ppm; ^{13}C {^{1}H} NMR ($D_2O/CD_3(CO)CD_3$, 1:3, 125.6 MHz): δ = 13.7 (s, CH_3), 13.8 (s, CH_3), 22.4 (s, CH_3CH_2), 22.6 (s, C_o), 25.6 (s, C_b), 26.6 (s, C_c), 29.1 (s, C_dH_2), 29.5 (br s, $C_{e\text{-}m}$), 31.2 (d, $^2J_{PC}$ = 16.2 Hz, NCH_3), 31.8 (d, $^3J_{PH}$ = 16.2 Hz, $CHCH_2$), 31.9 (s, C_n), 34.7 (s, $ArCH_2$), 39.7 (d, $^1J_{PC}$ = 75.5 Hz, PCH), 41.0 (s, $NHCH_2$), 48.1 (s, C_a), 49.9 (s, $C_{a'}$), 61.5 (s, CH_2OH), 62.1 (s, CH_2OH), 67.9 (s, CHOH), 69.0 (s, CHOH), 70.8 (s, CHOH), 71.2 (s, CHOH), 72.9 (s, CHOH), 75.6 (s, CHOH), 78.7 (s, CHO), 103.1 (s, C_{anom}), 121.1 (s, C_1^2), 121.2 (s, C_0^2), 128.3 (s, C_0^3), 129.8 (s, C_1^3), 132.6 (s, C_0^4), 136.5 (s, C_1^4), 138.9 (br s, CH=N), 149.1 (d, $^2J_{PC}$ = 6.9 Hz, C_1^1), 151.0 (br s, C_0^1), 168.9 (s, CONH) ppm.

3d-G1: Dendrimer **3b-G1** (50 mg, 0.008 mmol) was added to a solution of *N*-hexadecylaminolactitol (55 mg, 0.096 mmol) in distilled water (10 mL). After 3 days stirring at rt, the colorless homogeneous solution was freeze-dried to afford **3d-G1** as a white powder (104 mg, 100%). ^{31}P {^{1}H} NMR (D_2O/CD_3CN, 3:1, 121.5 MHz): δ = 9.2 (s, N_3P_3), 17.3 (s, PO_3Me_2), 62.8 (s, P=S) ppm; ^{1}H NMR (CD_3OD, 125.6 MHz): δ = 0.86 (br s, 36H, CH_3), 0.93 (br s, 36H, CH_3), 1.23–1.49 (m, 504H, CH_2 and $C_{c\text{-}o}H$), 1.51–1.68 (m, 24H, C_bH), 1.80 (br s, 24H, $ArCH_2$), 2.08–2.09 (m, 24H, $CHCH_2$), 2.51–2.67 (m, 12H, CHP), 2.73–2.92 (m, 18H, NCH_3), 3.11 (br s, 48H, C_aH_2 and $C_{a'}H_2$), 3.12–3.49 (m, 24H, $NHCH_2$), 3.55–3.99 (m, 156H, CHOH, CH_2OH and CH_{anom}), 6.81–7.93 (m, 12H, C_0^2H), 7.03–7.38 (m, 48H, C_1^2H and C_1^3H), 7.64 (br s, 12H, C_0^3H), 7.71 (br s, 6H, CH=N) ppm; ^{13}C{^{1}H} NMR (D_2O/CD_3COCD_3, 1:3, 125.6 MHz): δ = 13.7 (s, CH_3), 13.9 (s, CH_3), 22.1 (s, C_o), 22.3 (s, CH_2), 26.0 (s,

C_b), 26.6 (s, C_c), 28.3 (br s, CHCH$_2$), 29.3 (s, C_dH_2), 29.5 (br s, CH_2 and $C_{e\text{-}m}$), 31.7 (s, CH_2), 31.9 (s, C_n), 34.5 (s, ArCH$_2$), 37.5 (d, $^1J_{PC}$ = 75.5 Hz, PCH), 40.6 (s, NHCH$_2$), 48.1 (s, C_a), 50.0 (s, $C_{a'}$), 61.4 (s, CH$_2$OH), 62.1 (s, CH$_2$OH), 68.1 (s, CHOH), 69.0 (s, CHOH), 70.7 (s, CHOH), 71.2 (s, CHOH), 72.8 (s, CHOH), 75.6 (s, CHOH), 78.9 (s, CHO), 103.1 (s, C_{anom}), 121.1 (br s, C_0^2 and C_1^2), 128.0 (br s, C_0^3), 129.8 (s, C_1^3), 134.5 (s, C_0^4), 136.8 (s, C_1^4), 139.1 (s, CH=N), 149.2 (s, C_1^1), 152.3 (s, C_0^1), 173.5 (s, CONH) ppm.

4d-G1: Dendrimer **4b-G1** (50 mg, 0.013 mmol) was added to a solution of *N*-hexadecylaminolactitol (88 mg, 0.156 mmol) in distilled water (10 mL). After 3 days stirring at rt, the colorless homogeneous solution was freeze-dried to afford **4d-G1** as a white powder (137 mg, 100%). ^{31}P {^{1}H} NMR (D_2O/CD_3CN, 3:1, 121.5 MHz): δ = 9.1 (s, N_3P_3), 11.0 (s, PO_3Me_2), 62.8 (s, P=S) ppm; ^{1}H NMR (D_2O/CD_3COCD_3, 1:3, 500.1 MHz): δ = 0.98 (br s, 36H, CH_3), 1.18 (br s, 312 H, $C_{c-o}H_2$), 1.69 (br s, 24H, C_bH_2), 2.64–2.93 (m, 84H, Ar*CH*$_2$, PCH$_2$, C_aH and $C_{a'}$H), 3.33–3.49 (m, 42H, NH*CH*$_2$ and NCH_3), 3.41–4.15 (m, 120H, CHOH and CH_2OH) 4.15 (br s, 12H, CH_{anom}), 6.29 (br s, 12H, CHP), 7.00 (br s, 36H, C_0^2H and C_1^2H), 7.35 (br s, 24H, C_1^3H), 7.53 (br s, 12H, C_0^3H), 7.67 (br s, 6H, CH=N) ppm; ^{13}C {^{1}H} NMR (D_2O/CD_3CN, 3:1, 100.6 MHz): δ = 14.1 (s, CH_3), 22.9 (s, C_o), 26.2 (s, C_b), 27.0 (s, C_c), 29.8 (br s, C_d), 30.3 (br s, $C_{e\text{-}m}$), 32.3 (br s, C_n), 32.8 (br s, CH_3N), 48.2 (br s, C_a), 50.3 (br s, $C_{a'}$), 61.4 (s, CH_2OH), 62.8 (s, CH_2OH), 67.3 (s, CHOH), 69.0 (br s, CHOH), 70.6 (br s, CHOH), 71.3 (s, CHOH), 72.9 (br s, CHOH), 75.4 (br s, CHOH), 80.1 (br s, CHO), 103.2 (br s, C_{anom}), 121.6 (br s, C_0^2 and C_1^2), 124.5 (s, CHP), 126.4 (s, C_0^3), 128.5 (s, C_1^3), 132.6 (s, C_0^4), 134.7 (br s, C_1^4), 139.4 (br s, ArCH, CH=N), 147.7 (br s, C_1^1), 150.8 (s, C_0^1) ppm.

5d-G1: Dendrimer **5b-G1** (50 mg, 0.013 mmol) was added to a solution of *N*-hexadecylaminolactitol (88 mg, 0.156 mmol) in distilled water (10 mL). After 3 days stirring at rt, the colorless homogeneous solution was freeze-dried to afford **5d-G1** as a white powder (138 mg, 100%). ^{31}P {^{1}H} NMR (D_2O/CD_3CN, 3:1, 121.5 MHz): δ = 8.6 (s, N_3P_3), 15.9 (s, PO_3Me_2), 63.5 (s, P=S) ppm; ^{1}H NMR (D_2O/$CD_3(CO)CD_3$, 1:3, 500.1 MHz): δ = 1.02 (br s, 36H, CH_3), 1.39 (br s, 336 H, $C_{b\text{-}o}H_2$), 2.59–3.19 (m, 48H, C_aH and $C_{a'}$H), 3.36–3.63 (m, 18H, NCH_3), 3.65–4.29 (m, 156H, CHOH, CH_2OH and CHOHP), 7.31 (br s, 36H, C_0^2H and C_1^2H), 7.60 (br s, 24H, C_1^3H), 7.81 (br s, 18H, C_0^3H and CH=N) ppm; ^{13}C {^{1}H} NMR (D_2O/CD_3CN, 3:1, 75.5 MHz): δ = 14.1 (s, CH_3), 22.9 (s, C_o), 25.9 (s, C_b), 27.0 (s, C_c), 29.8 (s, C_d), 30.3 (br s, $C_{e\text{-}m}$), 32.3 (br s, C_n), 32.8 (br s, CH_3N), 46.7 (s, C_a), 48.3 (br s, $C_{a'}$), 61.4 (br s, CH_2OH), 62.5 (br s, CH_2OH), 68.0 (s, CHOH), 69.1 (br s, CHOH), 71.3 (br s, CHOH and CHOHP), 72.8 (br s, CHOH), 75.5 (br s, CHOH), 80.0 (br s, CHO), 103.4 (br s, CH_{anom}), 120.7 (br s, C_0^2 and C_1^2), 128.8 (br s, C_0^3 and C_1^3), 132.3 (br s, CH=N), 138.7 (br s, C_1^4), 140.4 (s, C_0^4), 149.3 (br s, C_1^1), 151.1 (br s, C_0^1) ppm.

6d-G1: Dendrimer **6b-G1** (50 mg, 0.009 mmol) was added to a solution of *N*-hexadecylaminolactitol (123 mg, 0.216 mmol) in distilled water (10 mL). After 3 days stirring at rt, the colorless homogeneous solution was freeze-dried to afford **6d-G1** as a white powder (170 mg, 100%). ^{31}P {^{1}H} NMR (D_2O/CD_3CN, 3:1, 121.5 MHz): δ = 8.6 (s, N_3P_3), 7.2 (s, PO_3Me_2), 63.0 (s, P=S) ppm; ^{1}H NMR (D_2O/CD_3COCD_3, 1:3, 500.1 MHz): δ = 0.93 (br s, 72H, CH_3), 1.32 (br s, 624H, $C_{c-o}H_2$), 1.73 (br s, 48H, C_bH_2), 2.82–3.06 (m, 96H, C_aH and $C_{a'}$H), 3.24–3.46 (m, 66H, NCH_2P and NCH_3), 3.63–4.20 (m, 288H, CHOH and CH_2OH), 4.51 (br s, 24H, CH_{anom}), 6.89–7.12 (m, 12H, C_0^2H), 7.20 (br s, 24H, C_1^2H), 7.40 (br s, 12H, C_1^3H), 7.73 (br s, 12H, C_0^3H), 7.89 (br s, 6H, CH=N) ppm; ^{13}C {^{1}H} NMR (D_2O/CD_3CN, 3:1, 125.8 MHz): δ = 14.0 (s, CH_3), 22.8 (s, C_o), 26.0 (s, C_b), 26.8 (s, C_c), 29.7 (s, C_d), 30.2 (br s, $C_{e\text{-}m}$ and Ar*CH*$_2$), 32.2 (s, C_n), 32.7 (br s, CH_3N), 47.9 (br s, C_a), 50.1 (br s, $C_{a'}$), 53.1 (br s, NCH_2P), 57.4 (br s, CH_2N), 61.3 (s, CH_2OH), 62.2 (br s, CH_2OH), 67.8 (br s, CHOH), 68.8 (br s, CHOH), 71.1 (br s, CHOH), 72.8 (br s, CHOH), 75.3 (br s, CHOH), 77.2 (br s, CHOH), 79.7 (br s, CHO), 103.2 (br s, C_{anom}), 121.5 (br s, C_0^2 and C_1^2), 128.3 (br s, C_0^3), 130.6 (br s, C_1^3), 132.5 (br s, C_0^4), 134.5 (br s, C_1^4), 139.2 (br s, CH=N), 149.6 (br s, C_1^1), 151.0 (br s, C_0^1) ppm.

4.2. Antiviral assays

CEM-SS cells were cultured in RPMI medium supplemented with 10% fetal calf serum (heated at 56 °C for 30 min). CEM-SS cells were infected with HIV-1 LAI and the production of virus was evaluated after five days, by measuring the reverse transcriptase (RT) which expresses the presence of HIV in the supernatant culture medium. RT inhibition percentage, providing IC_{50} values (concentration of drug at which virus production is inhibited by 50%), was determined in comparison with the non-treated cells. The cytotoxicities evaluation is based on the viability of the non-infected cells using a colorimetric assay. This colorimetric MTT test is based on the capacity of living cells to reduce MTT to formazan. The produced quantity of formazan (characterized by OD540) is directly proportional to the number of living cells and to the CC_{50} (concentration at which OD540 was reduced by half).

Acknowledgments

The European Community is acknowledged for funding support (A. P.-A., Marie Curie EST 'Nanotool').

Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmc.2009.10.058.

References and notes

1. Buhleier, E.; Wehner, F.; Vögtle, F. *Synthesis* **1978**, *78*, 155.
2. Rolland, O.; Turrin, C.-O.; Caminade, A.-M.; Majoral, J.-P. *New J. Chem.* **2009**, *33*, 1809.
3. Patri, A. K.; Kukowska-Latallo, J. F.; Baker, J. R. *Adv. Drug Delivery Rev.* **2005**, *57*, 2203.
4. Blanzat, M.; Turrin, C. O.; Aubertin, A. M.; Couturier-Vidal, C.; Caminade, A. M.; Majoral, J. P.; Rico-Lattes, I.; Lattes, A. *ChemBioChem* **2005**, *6*, 2207.
5. Najlah, M.; D'Emanuele, A. *Curr. Opin. Drug Discovery Dev.* **2007**, *10*, 756.
6. Myc, A.; Douce, T. B.; Ahuja, N.; Kotlyar, A.; Kukowska-Latallo, J.; Thomas, T. P.; Baker, J. R. *Anti-Cancer Drugs* **2008**, *19*, 143.
7. Rosa Borges, A.; Schengrund, C. L. *Curr. Drug Targets Infect. Disord.* **2005**, *5*, 247.
8. Mammen, M.; Choi, S. K.; Whitesides, G. M. *Angew. Chem., Int. Ed.* **1998**, *37*, 2754–2794.
9. Kitov, P. I.; Bundle, D. R. *J. Am. Chem. Soc.* **2003**, *125*, 16271.
10. Dutta, T.; Garg, M.; Jain, N. K. *Eur. J. Pharm. Sci.* **2008**, *34*, 181.
11. Dutta, T.; Jain, N. K. *Biochim. Biophys. Acta, Gen. Subj.* **2007**, *1770*, 681.
12. Weber, N.; Ortega, P.; Clemente, M. I.; Shcharbin, D.; Bryszewska, M.; de la Mata, F. J.; Gomez, R.; Munoz-Fernandez, M. A. *J. Controlled Release* **2008**, *132*, 55.
13. Berger, E. A.; Murphy, P. M.; Farber, J. M. *Annu. Rev. Immunol.* **1999**, *17*, 657.
14. Kensinger, R. D.; Catalone, B. J.; Krebs, F. C.; Wigdahl, B.; Schengrund, C. L. *Antimicrob. Agents Chemother.* **2004**, *48*, 1614.
15. Kensinger, R. D.; Yowler, B. C.; Benesi, A. J.; Schengrund, C. L. *Bioconjugate Chem.* **2004**, *15*, 349.
16. Wang, S. K.; Liang, P. H.; Astronomo, R. D.; Hsu, T. L.; Hsieh, S. L.; Burton, D. R.; Wong, C. H. *Proc. Natl. Acad. Sci. U.S.A.* **2008**, *105*, 3690.
17. Yahi, N.; Sabatier, J.-M.; Nickel, P.; Mabrouk, K.; Gonzalez-Scarano, F.; Fantini, J. *J. Biol. Chem.* **1994**, *269*, 24349.
18. Alfsen, A.; Bomsel, M. *J. Biol. Chem.* **2002**, *277*, 25649.
19. Faroux-Corlay, B.; Greiner, J.; Terreux, R.; Cabrol-Bass, D.; Aubertin, A. M.; Vierling, P.; Fantini, J. *J. Med. Chem.* **2001**, *44*, 2188.
20. Rico-Lattes, I.; Blanzat, M.; Franceschi-Messant, S.; Perez, E.; Lattes, A. *C.R. Chimie* **2005**, *8*, 807.
21. Rico-Lattes, I.; Garrigues, J. C.; Perez, E.; André-Barrès, C.; Madelaine-Dupuich, C.; Lattes, A.; Linas, M. D.; Aubertin, A. M. *New J. Chem.* **1995**, *19*, 341.
22. Moulard, M.; Lortat-Jacob, H.; Mondor, I.; Roca, G.; Wyatt, R.; Sodroski, J.; Zhao, L.; Olson, W.; Kwong, P. D.; Sattentau, Q. J. *J. Virol.* **2000**, *74*, 1948.
23. McCarthy, T. D.; Karellas, P.; Henderson, S. A.; Giannis, M.; O'Keefe, D. F.; Heery, G.; Paull, J. R. A.; Matthews, B. R.; Holan, G. *Mol. Pharm.* **2005**, *2*, 312.
24. Weber, J.; Desai, K.; Darbyshire, J. *Plos Med.* **2005**, *2*, 392.
25. McCarthy, T.; Karellas, P.; Henderson, S.; Giannis, M.; O'Keefe, D.; Matthews, B.; Braggs, B.; Paull, J.; Heery, G.; Krippner, G.; Holan, G. *Antiviral Res.* **2004**, *62*, A44.

26. Blanzat, M.; Turrin, C. O.; Perez, E.; Rico-Lattes, I.; Caminade, A. M.; Majoral, J. P. *Chem. Commun.* **2002**, 1864.
27. Iampietro, D. J.; Kaler, E. W. *Langmuir* **1999**, *15*, 8590.
28. Consola, S.; Blanzat, M.; Perez, E.; Garrigues, J. C.; Bordat, P.; Rico-Lattes, I. *Chem. Eur. J.* **2007**, *13*, 3039.
29. Pasc-Banu, A.; Stan, R.; Blanzat, M.; Perez, E.; Rico-Lattes, I.; Lattes, A.; Labrot, T.; Oda, R. *Colloids Surf., A* **2004**, *242*, 195.
30. Soussan, E.; Mille, C.; Blanzat, M.; Bordat, P.; Rico-Lattes, I. *Langmuir* **2008**, *24*, 2326.
31. Caminade, A. M.; Maraval, V.; Laurent, R.; Turrin, C. O.; Sutra, P.; Leclaire, J.; Griffe, L.; Marchand, P.; Baudoin-Dehoux, C.; Rebout, C.; Majoral, J. P. *C.R. Chimie* **2003**, *6*, 791.
32. Poupot, M.; Griffe, L.; Marchand, P.; Maraval, A.; Rolland, O.; Martinet, L.; L'Faqihi-Olive, F. E.; Turrin, C. O.; Caminade, A. M.; Fournié, J. J.; Majoral, J. P.; Poupot, R. *FASEB J.* **2006**, *20*, 2339.
33. Marchand, P.; Griffe, L.; Poupot, M.; Turrin, C.-O.; Bacquet, G.; Fournié, J.-J.; Majoral, J.-P.; Poupot, R.; Caminade, A.-M. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 3963.
34. Blais, J. C.; Turrin, C. O.; Caminade, A. M.; Majoral, J. P. *Anal. Chem.* **2000**, *72*, 5097.
35. Pérez-Anes, A.; Spataro, G.; Coppel, Y.; Blanzat, M.; Turrin, C.-O.; Moog, C.; Caminade, A.-M.; Rico-Lattes, I.; Majoral, J.-P. *Org. Biomol. Chem.* **2009**, *7*, 3491.
36. Moog, C.; Wick, A.; Ber, P. L.; Kirn, A.; Aubertin, A.-M. *Antiviral Res.* **1994**, *24*, 275.
37. Pauwels, R.; Balzarini, J.; Baba, M.; Snoeck, R.; Schols, D.; Herdewijn, P.; Desmyter, J.; Clercq, E. D. *J. Virol. Methods* **1988**, *20*, 309.
38. Peytou, V.; Condom, R.; Patino, N.; Guedj, R.; Aubertin, A.-M.; Gelus, N.; Bailly, C.; Terreux, R.; Cabrol-Bass, D. *J. Med. Chem.* **1999**, *42*, 4042.
39. Fletcher, P. S.; Wallace, G. S.; Mesquita, P. M. M.; Shattock, R. J. *Retrovirology* **2006**, *3*, 46.
40. Macri, R. V.; Karlovská, J.; Doncel, G. F.; Du, X.; Maisuria, B. B.; Williams, A. A.; Sugandhi, E. W.; Falkinham, J. O., III; Esker, A. R.; Gandour, R. D. *Bioorg. Med. Chem.* **2009**, *17*, 3162.
41. Fichorova, R. N.; Tucker, L. D.; Anderson, D. J. *J. Infect. Dis.* **2001**, *184*, 418.
42. Rolland, O.; Griffe, L.; Poupot, M.; Maraval, A.; Ouali, A.; Coppel, Y.; Fournié, J. J.; Bacquet, G.; Turrin, C. O.; Caminade, A. M.; Majoral, J. P.; Poupot, R. *Chem. Eur. J.* **2008**, *14*, 4836.
43. Perrin, D. D.; Armarego, W. L. F. *Purification of Laboratory Chemicals*, 3rd ed.; Pergamon Press: Oxford, 1988.
44. Launay, N.; Caminade, A. M.; Lahana, R.; Majoral, J. P. *Angew. Chem., Int. Ed. Engl.* **1994**, *33*, 1589.
45. Launay, N.; Caminade, A. M.; Majoral, J. P. *J. Organomet. Chem.* **1997**, *529*, 51.
46. Vivares, D.; Soussan, E.; Blanzat, M.; Rico-Lattes, I. *Langmuir* **2008**, *24*, 9260.