Identification of a Terphenyl Derivative that Blocks the Cell Cycle in the G_0-G_1 Phase and Induces Differentiation in Leukemia Cells

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To further explore the SAR of resveratrol-related *trans*-stilbene derivatives, here we describe the synthesis of (a) a series of 3,5-dimethoxy analogues in which a variety of substituents were introduced at positions 2′, 3′, 4′, and 5′ of the stilbene scaffold and (b) a second group of derivatives (2-phenylnaphthalenes and terphenyls) that incorporate a phenyl ring as a bioisosteric replacement of the stilbene alkenyl bridge. We thoroughly characterized all of the new compounds with respect to their apoptosis-inducing activity and their effects on the cell cycle. One of the new derivatives, **13g**, behaved differently from the others, as it was able to block the cell cycle in the G_0-G_1 phase and also to induce differentiation in acute myelogenous leukemia HL60 cells. Compared to resveratrol, the synthetic terphenyl **13g** showed a more potent apoptotic and differentiating activity. Moreover, it was active on both multidrug resistance and Bcr-Abl-expressing cells that were resistant to resveratrol.

Introduction

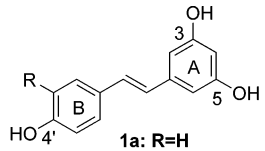
The stilbene scaffold is a basic element for a number of biologically active natural and synthetic compounds, and in accordance with Evans' definition, it can be considered as a privileged structure.[1] One of the most relevant and studied stilbenes is resveratrol (**1a**, 3,4′,5-trihydroxy-*trans*-stilbene; Chart 1), a phytoalexin present in grapes and other food products,[2,3] endowed with several different biological properties and able to act and bind at different cellular targets.[4,5]

The interesting chemopreventive[6,7] and chemotherapeutic[7−11] properties associated with resveratrol offer promises for the design of new chemoterapeutic agents, and in this context, several studies have been reported describing the synthesis and antitumor evaluation of these types of stilbene derivatives.[12−16]

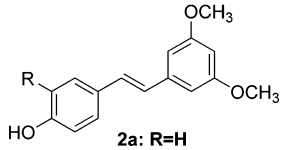
Resveratrol has also been shown to induce apoptosis in different cancer cell lines.[7,17−19] Since reduced apoptosis has been implicated in the development and progression of malignant tumors[20,21] and in the occurrence of chemoresistant phenotypes,[22−25] resveratrol-induced apoptosis might therefore contribute to its antitumor activity. However, resveratrol is not a potent cytotoxic compound when compared with other chemotherapeutic drugs. Recently, we observed in different cancer cell lines that, to obtain the induction of apoptosis in 50% of cells, a resveratrol concentration about 10 times higher than its IC_{50} was necessary.[26]

Because of our ongoing interest in finding new apoptosis-inducing agents, we are particularly intrigued by molecular scaffolds that allow the parallel synthesis of substituted derivatives. Actually, the parallel procedure is suitable for rapidly obtaining variously substituted analogues, particularly when structure-based design strategy is not applicable due to the lack of knowledge of a specific target. Thus, we recently synthesized a small library of resveratrol analogues bearing the 3,5-dimethoxy motif at the A phenyl ring and amino, methoxy, and hydroxy functions at the 3′- and/or 4′-positions.[26] Many derivatives turned out to be more active than resveratrol as apoptosis-inducing agents in HL60 leukemia cells. Besides, some of them were active toward resistant HL60R cells, and their activity was higher than that of several classic chemotherapeutic agents. Also, considering the importance of the 3,5-dimethoxy motif in conferring proapoptotic activity to stilbene-based compounds,

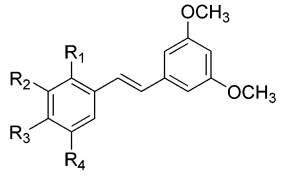
Chart 1[a]



1a: R=H
1b: R=OH



2a: R=H
2b: R=OH



3a: R_1=OH, R_2=R_3=R_4=H (55%)[a]
3b: R_1=R_3=R_4=H, R_2=OH (50%)[a]
3c: R_1=R_2=R_4=H, R_3=OCH_3 (70%)[a]
3d: R_1=OH, R_2=OCH_3, R_3=R_4=H (50%)[a]
3e: R_1=R_4=H, R_2=OCH_2CH_3, R_3=OH (45%)[a]
3f: R_1=OH, R_2=R_3=H, R_4=Cl (50%)[a]
3g: R_1=H, R_2=R_4=OCH_3, R_3=OH (45%)[a]
3h: R_1=H, R_2=R_4=$C(CH_3)_3$, R_3=OH (40%)[a]

[a] Overall yield.

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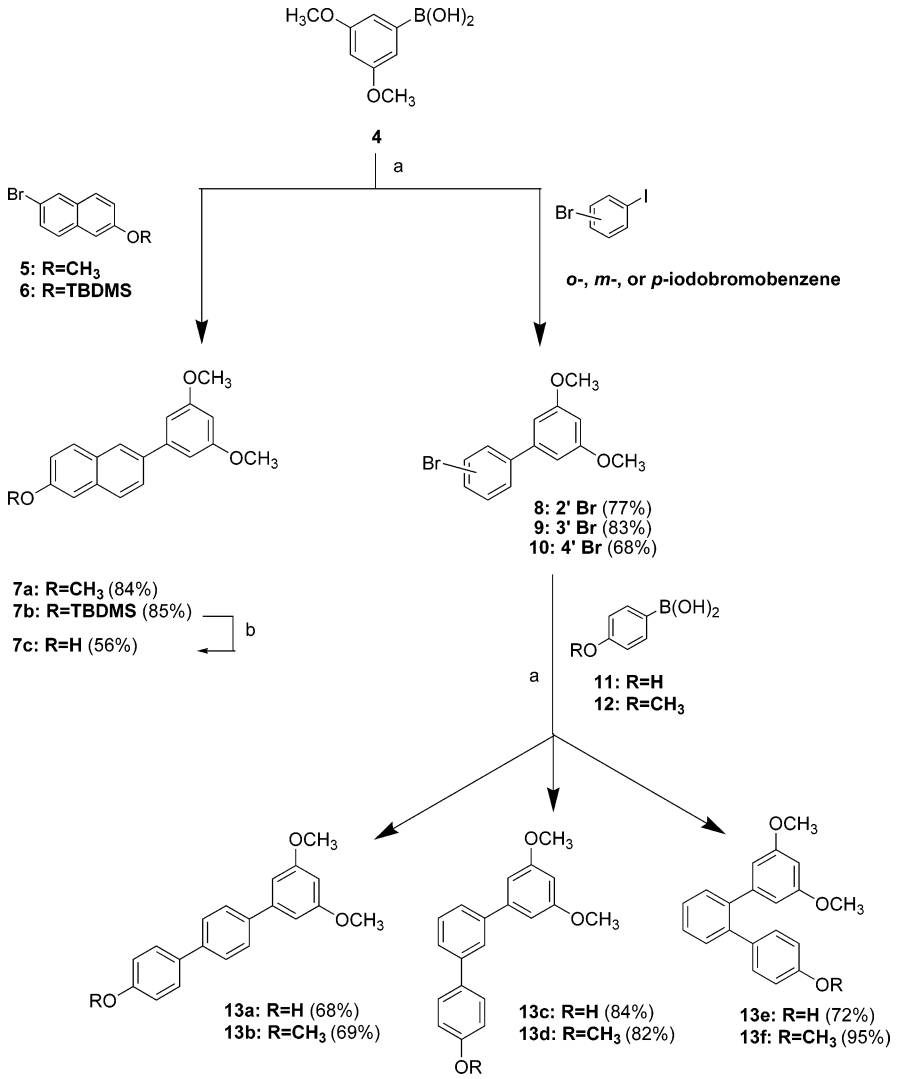
Scheme 1[a]



[a] Reagents and conditions: (a) $Pd(Ph_3P)_4$, aq Na_2CO_3, toluene/EtOH 3:1, reflux, 5 h; Bu_4NF, THF, rt, 3 h.

we compared the cellular effects of pterostilbene (**2a**) and 3′-hydroxypterostilbene (**2b**) with those of their corresponding natural 3,5-hydroxy analogues, resveratrol (**1a**) and piceatannol (**1b**), respectively.[27] The 3,5-dimethoxylated compounds differed with respect to **1a** and **1b** not only in their pattern of activity on sensitive and resistant cells but also regarding the effects on the cell cycle and apoptotic pathways. Moreover, both **2a** and **2b** were less toxic to normal hemopoietic stem cells than to the leukemia and lymphoma cells.

To further explore the SAR of this class of derivatives, here we describe the synthesis of a series of resveratrol analogues (**3a**−**h**, Chart 1) in which a variety of substituents were introduced at positions 2′, 3′, 4′, and 5′ of the stilbene scaffold while the 3,5-dimethoxy motif was maintained. We thoroughly characterized all of the new derivatives with respect to both their apoptosis-inducing activity and their effects on cell cycle by carrying out experiments on leukemia cell lines HL60, HL60R (MDR expressing), and K562 (apoptosis resistant). Some of the compounds turned out to be active as apoptosis-inducing agents; however, they did not reveal significant profile variations in terms of cytotoxicity and effects on cell cycle with respect to resveratrol and our previous stilbene derivatives.[26]

Given these results, we aimed at increasing the structural diversity of the new molecules by synthesizing a second series of derivatives, which incorporate a phenyl ring as a bioisosteric substitution of the alkenyl bridge. Thus, we obtained both 2-phenylnaphthalenes **7a**,**c**,**d** (Schemes 1 and 2), and terphenyls **13a**−**i** (Schemes 1 and 2), all of which present the resveratrol-like pattern of oxygenated phenyl rings, i.e., one para-substituted and one di-meta-substituted phenyl connected by an unsaturated/aromatic system. One of these compounds, **13g**, behaved differently than the others, as it was able to block the cell cycle in the G_0-G_1 phase and also to induce differentiation in HL60 cells.

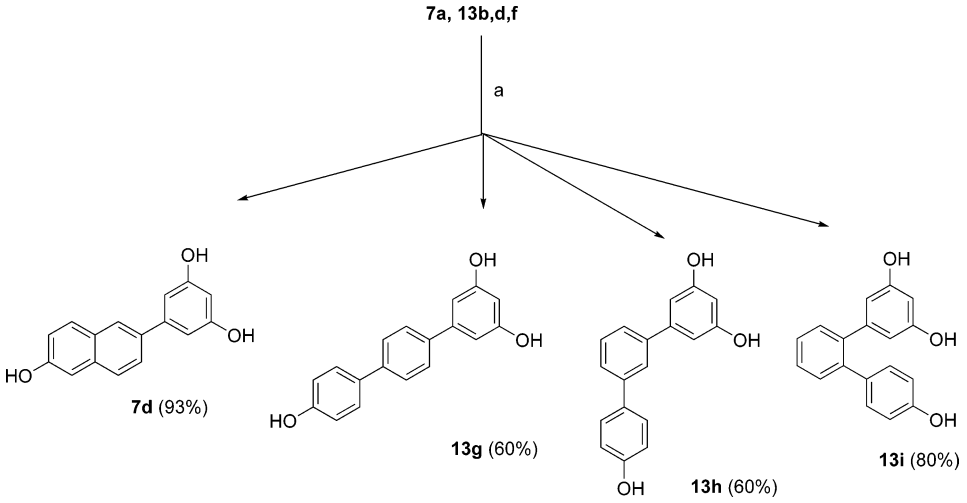
Chemistry

The compounds were obtained following different parallel synthetic pathways (Schemes 1 and 2) carried out in a Carousel reaction station.

To prepare the *trans*-stilbene derivatives **3a**−**h** (Chart 1), we took advantage of a standard Wittig reaction between the appropriate aromatic aldehydes and the aromatic ylide[26,28] (see the Supporting Information).

The 2-phenylnaphthalene derivatives and the terphenyls were obtained as shown in Scheme 1. A cross-coupling Suzuki reaction between 3,5-dimethoxyphenylboronic acid **4**[29a,b] and the appropriate 2-bromonaphthalene derivatives **5**, **6**, or bromoiodobenzene (ortho, meta, or para isomers) in the presence of a catalytic amount of tetrakis(triphenylphosphine)palladium

Scheme 2[a]



[a] Reagents and conditions: (a) BBr_3, CH_2Cl_2, −78 °C, 18 h.

Table 1. IC_{50} (μM ± SE) and AC_{50} (μM ± SE) Values of Resveratrol (**1a**), Pterostilbene (**2a**), Stilbene Derivatives **3a**−**h**, 2-Phenylnaphthalenes **7a,c,d**, and Terphenyls **13a**−**i** in Sensitive HL60, MDR HL60R, and Bcr-Abl-Expressing K562 Cells

	HL60		HL60R		K562	
compounds	IC_{50}[a]	AC_{50}[b]	IC_{50}[a]	AC_{50}[b]	IC_{50}[a]	AC_{50}[b]
resveratrol (**1a**)	5.0 ± 0.8	50 ± 6	60 ± 7	>100	28 ± 2	>100
pterostilbene (**2a**)	35 ± 4	75 ± 5	42 ± 3	85 ± 11	10 ± 3	45 ± 7
3a	9.0 ± 0.6	35 ± 4	68 ± 7	>100	24 ± 2	72 ± 8
3b	23 ± 5	58 ± 5	>100	>100	26 ± 1	70 ± 13
3c	2.5 ± 0.3	4.0 ± 0.3	8.0 ± 0.9	50 ± 2	10 ± 1	25 ± 4
3d	3.5 ± 0.4	15 ± 2	34 ± 5	82 ± 10	8.0 ± 0.8	28 ± 2
3e	11 ± 2	18 ± 2	42 ± 6	98 ± 10	18 ± 3	60 ± 9
3f	14 ± 2	29 ± 5	73 ± 8	>100	16 ± 1	55 ± 7
3g	20 ± 2	79 ± 6	>100	>100	23 ± 4	75 ± 5
3h	84 ± 6	>100	>100	>100	>100	>100
7a	76 ± 3	91 ± 18	>100	>100	>100	>100
7c	10 ± 1	19 ± 1	16 ± 2	33 ± 6	42 ± 5	55 ± 4
7d	28 ± 4	48 ± 7	53 ± 3	92 ± 15	20 ± 4	84 ± 13
13a	2.5 ± 0.2	30 ± 3	23 ± 3	32 ± 4	35 ± 5	42 ± 5
13b	75 ± 7	>100	>100	>100	>100	>100
13c	4.8 ± 0.7	23 ± 2	20 ± 2	48 ± 2	14 ± 2	27 ± 3
13d	50 ± 7	>100	80 ± 12	>100	10 ± 2	50 ± 5
13e	5.0 ± 0.3	10 ± 3	28 ± 2	24 ± 4	14 ± 2	18 ± 3
13f	7.0 ± 0.4	15 ± 2	16 ± 2	48 ± 5	6.0 ± 0.8	8.0 ± 0.7
13g	7.0 ± 1.2	25 ± 3	23 ± 4	60 ± 9	20 ± 2	75 ± 12
13h	27 ± 3	63 ± 5	58 ± 9	>100	20 ± 2	75 ± 9
13i	3.5 ± 0.3	9.0 ± 0.7	12 ± 3	40 ± 3	8.0 ± 0.6	38 ± 4

[a] Concentration able to inhibit 50% of cell growth. [b] Concentration able to induce apoptosis in 50% of cells.

gave the 2-phenylnaphthalene derivatives **7a**,**b** and the biphenyl dimethoxy derivatives **8**−**10**, respectively. A second Suzuki cross-coupling between compounds **8**−**10** and the boronic acids **11** and **12** provided the required terphenyls **13a**−**f**. Removal of the *tert*-butyldimethylsilyl (TBDMS) group from derivative **7b** using tetrabutylammonium fluoride afforded compound **7c**. Finally, the parallel demethylation of trimethoxy derivatives **7a** and **13b**,**d**,**f** with BBr_3 afforded **7d** and **13g**−**i** (Scheme 2). Compounds **7a**,**c**,**d** were recently described by Minutolo et al.[30] within a series of resveratrol analogues tested against human breast cancer cells.

Biology

All compounds considered in this study were tested for their antiproliferative and proapoptotic activities on sensitive acute myelogenous leukemia HL60 cells, multidrug resistance (MDR)-expressing HL60R cells, and Bcr-Abl-expressing K562 cells.

The antiproliferative activity of each compound was evaluated by counting cells with an automatic cell counter; apoptosis was evaluated by morphological assay and annexine V test. The effects of each compound on cell cycle were studied by flow cytometry after staining cells with propidium iodide. Moreover, for compounds able to block cells in the G_0−G_1 phase, we investigated the ability to induce cell differentiation by evaluating the expression of CD11b, CD11c, and CD14 on HL60 cells.

Results

The antiproliferative and apoptosis-inducing activities of the new compounds are summarized in Table 1, and these data are compared to those obtained with natural stilbenes resveratrol (**1a**) and its 3,5-dimethoxy analogue pterostilbene (**2a**). Antiproliferative and apoptosis-inducing activities were expressed as IC_{50} (concentration able to inhibit 50% of cell growth) and AC_{50} (concentration able to induce apoptosis in 50% of cells), respectively.

Within the 3,5-dimethoxystilbene series **3a**−**h**, the best results were obtained with compound **3c** bearing a 4′-methoxy function. The activity of 2′-hydroxy derivative **3a** was higher than that of pterostilbene (**2a**) on HL60 cells, but it became lower on HL60R and K562 cells. When the hydroxy function was moved

to the 3′ position, as in compound **3b**, the cytotoxic effects decreased on both HL60 and HL60R cells. In the subset of the B-ring disubstituted compounds, the introduction of the 2′-hydroxy and 3′-methoxy functions, as in compound **3d**, increased the proapoptotic activity on all cell lines tested in this study. The potency dropped by removing the 3′-methoxy group and introducing a 5′-Cl as in **3f**. By appending an ethoxy function in 3′-position of pterostilbene, we obtained compound **3e**, which showed a better activity on HL60 cells with respect to the parent compound **2a**. The analogue **3g** bearing 3′,5′-dimethoxy substitution was slightly less active than pterostilbene (**2a**), and the activity decreased further with the 3′,5′-di-*tert*-butyl analogue **3h**, which was the least active one of this series. In summary, these 3,5-dimethoxy-*trans*-stilbenes showed a cytotoxic profile comparable to that of other stilbene derivatives earlier synthesized by us,[26,27] even though some compounds of the previous series were more potent on HL60 cells.

Regarding the naphthalene derivatives **7a**,**c**,**d**, compound **7c**, which may be considered a pterostilbene analogue, showed an interesting activity on MDR-expressing HL60R cells, while the trimethoxy derivative **7a** was poorly active in all cell lines. The cytotoxic activity of **7d** was similar to that of the parent compound resveratrol (**1a**).

Among the terphenyl derivatives, compounds **13a**,**c**,**e**,**g** were effective antiproliferative and proapoptotic agents; in contrast, compounds **13b**,**d**,**h** showed a lower cytotoxic activity. Compounds **13f** and **13i** were the most potent compounds of the series.

Despite some evident effects of the structural changes on both the antiproliferative and proapoptotic actions of compounds, no clear SAR pattern could be identified throughout the series.

The effects of compounds on cell cycle were evaluated in HL60 cells by flow cytometry after staining cells with propidium iodide. Analysis of sub-G_0-G_1 (apoptotic peak), G_0-G_1, S, and G_2-M peaks revealed that all compounds presented in this study caused a recruitment of cells in S or G_2-M phases, except for the terphenyl **13g**, which blocked cells in G_0-G_1. In Figure 1b−f, the flow cytometric analyses of the most representative derivatives **3c**, **3d**, **7c**, **13f**, and **13i** are reported, compared to untreated HL60 cells (Figure 1a). Compound **3d** (Figure 1b) caused a clear recruitment of cells in S phase and a decrease of cells in the G_2-M phase; compounds **7c** and **13f** (Figure 1d,e) induced a recruitment of cells in S and G_2-M phases and a decrease of the G_0-G_1 peak, and compounds **3c** and **13i** (Figure 1c,f) caused a marked block of cells in the G_2-M phase. All compounds induced the appearance of a sub-G_0-G_1 apoptotic peak. Similar results were previously described for resveratrol, piceatannol, and other natural stilbenes.[27,31a−e] Figure 2b−d shows the effects of compound **13g** on cell cycle after 8 h (Figure 2b), 24 h (Figure 2c), and 96 h (Figure 2d). The block of cell cycle in G_0-G_1 is evident in Figure 2b−d when compared to the control (Figure 2a). An apoptotic sub-G_0-G_1 peak was observed in HL60 cells exposed to **13g** for 24 and 96 h (Figure 2c,d). In contrast, the sub-G_0-G_1 peak was very low in the sample exposed to **13g** for 8 h (Figure 2b).

Given the above results, although **13g** was not the most potent compound as regards the cytotoxic activity, we considered it worth further study, because it was the only one able to arrest the cell cycle in the G_0-G_1 phase. The G_0-G_1 recruitment appeared after 8 h when HL60 cells were exposed to 50 μM of **13g**; using a lower concentration (10 μM), we observed a G_0-G_1 block after 96 h (Figure 2d).

Considering the ability of **13g** to induce a stable block in the G_0-G_1 phase, we investigated its effect on cell differentiation

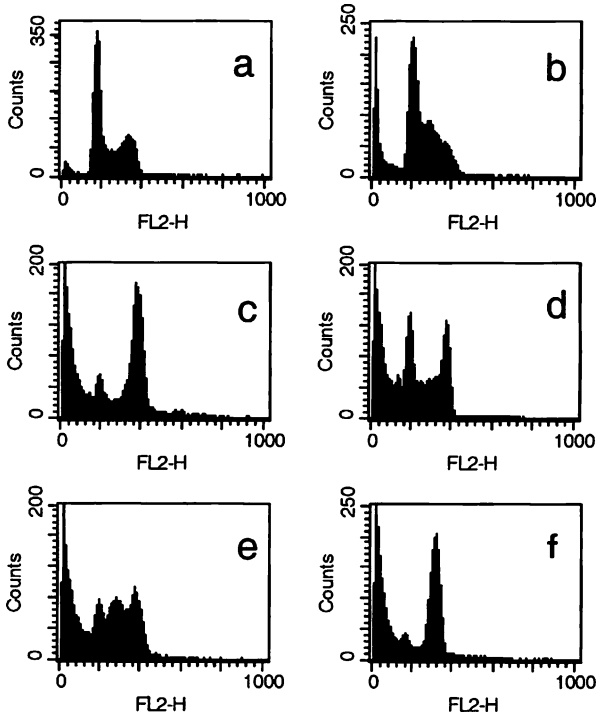


Figure 1. Cell cycle distribution of HL60 cells exposed for 24 h to some representative stilbene, naphthalene, and terphenyl compounds presented in this study: (a) control, (b) **3d** (30 μM), (c) **3c** (10 μM), (d) **7c** (20 μM), (e) **13f** (30 μM), (f) **13i** (20 μM).

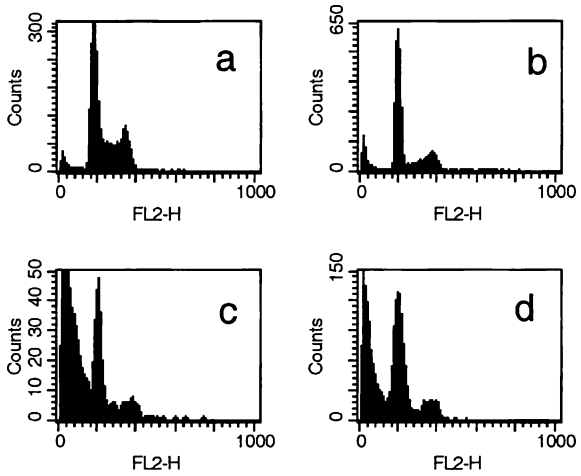


Figure 2. Cell cycle distribution of HL60 cells exposed for 8 h (b) and 24 h (c) to 50 μM of **13g**; (d) cell cycle distribution of HL60 cells exposed for 96 h to 10 μM of **13g**; (a) control.

in HL60 cells. Moreover, we compared the differentiating effect of **13g** with that of resveratrol (**1a**), because previous evidence indicated that resveratrol was able to induce cell differentiation in some promyelocytic leukemia cell lines.[32] Cells were exposed to 10 μM **13g** or 10 μM resveratrol. After 72 h, the expression of CD14 (monocytic marker), CD11b, and CD11c (granulocytic markers) was evaluated by flow cytometry. About 60% of HL60 cells exposed to **13g** became CD14, CD11b, and CD11c positive, while less than 25% of HL60 cells exposed to resveratrol became positive to these markers (Table 2). As shown in Figure 3a−f, HL60 cells differentiated in metamyelocytes, myelocytes, monocytes, and granulocytes after 4 days exposure to 10 μM **13g**. Of interest, some monocytes derived from HL60 cells exposed for 4 days to 10 μM **13g** became specialized macrophages able to phagocyte fragments of apoptotic cells (Figure 3f).

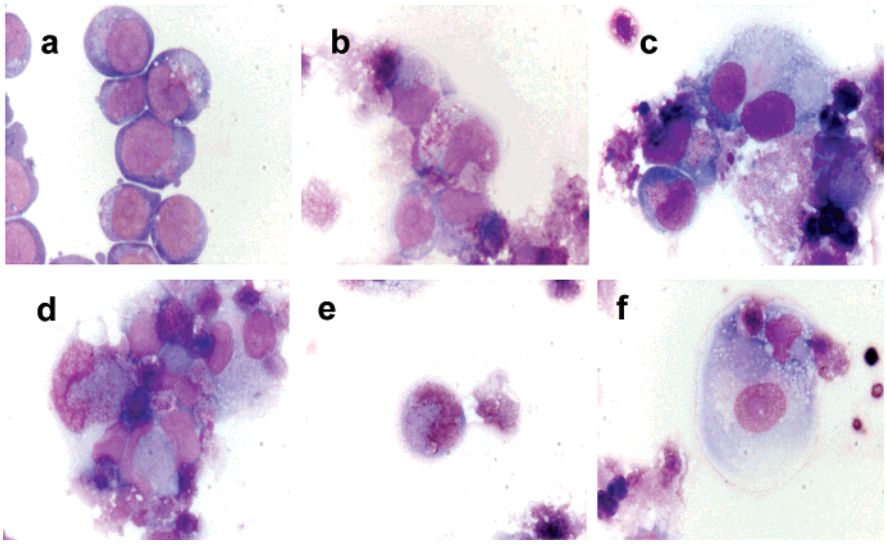


Figure 3. Morphologic changes observed in HL60 cells (promyelocytic acute leukemia) after 72−96 h exposure to **13g** (10 μM): (a) control (promyelocytes); (b) myelocytes and a metamyelocyte (72 h after exposure to **13g**); (c) myelocyte-, metamyelocyte-, and monoblast-derived cells (96 h after exposure); (d) metamyelocyte- and monoblast-derived cells (96 h after exposure); (e) band neutrophil (96 h after exposure); (f) phagocytosis of apoptotic bodies by a macrophage (96 h after exposure).

Table 2. Expression of Monocytic (CD14) and Granulocytic (CD11b, CD11c) Markers in HL60 Cells before (control) or 4 days after Treatment with 10 μM 13g or 10 μM Resveratrol (**1a**)

compounds	CD14 (%)	CD11b (%)	CD11c (%)
control	0.2	0	0
13g	58	62	60
resveratrol (**1a**)	23	24	22

In contrast, other trihydroxylated resveratrol-like compounds (**7d**, **13h**, **13i**) did not induce differentiation.

Discussion

As an extension of our previous studies[26,27] on the SAR of *trans*-stilbene derivatives, we synthesized a series of 3,5-dimethoxy analogues in which a variety of substituents were introduced at positions 2′, 3′, 4′, and 5′ of the stilbene scaffold. Moreover, in an attempt to increase the diversity of the compounds while the basic structural motif of resveratrol was retained, we obtained both some naphthalene and some terphenyl derivatives. Notably, the latter two structures do not bear the ethylene double bond that is the main reason for the chemical and metabolic instability of stilbene compounds.[33a−c] By the use of this approach, we earlier disclosed a terphenyl analogue of stilbene-based arotinoid that surprisingly supported cell growth and inhibited apoptosis.[34] The same strategy was also recently exploited by Minutolo et al., who carried out a study on the ceramide-mediated proapoptotic activity of some variously hydroxylated and methoxylated naphthalenes.[30]

All of the compounds presented in this paper were prepared through parallel synthesis, taking advantage of the standard Wittig reaction for stilbenes and of cross-coupling Suzuki protocol for naphthalenes and terphenyls. This synthetic approach proved to be efficient to rapidly obtain small sets of compounds and appropriate when random substitutions have to be explored in the lack of either a specific target or a clearly defined SAR pattern.

Initially, the stilbene compounds **3a**−**h** were studied in vitro for both cell growth inhibition and the ability to induce apoptosis in leukemia and lymphoma cells. In our previous work,[26] we pointed out the importance of a hydroxy function at the C-3′ position of the stilbene scaffold, and in a further development, we showed the high potency of 3′-hydroxypterostilbene (**2b**).[27] None of the variations presently introduced into the B ring of the compounds allowed us to improve the antiproliferative and proapoptotic profile of our lead **2b**. We studied not only sensitive HL60 cells but also resistant HL60R and Bcr-Abl-expressing K562 cells: despite some good potency showed by stilbene derivatives **3c** and **3d** on sensitive cells, the antiproliferative and eventually the proapoptotic actions were not maintained against resistant and Bcr-Abl-expressing cells. Among the naphthalene-based compounds **7a**,**c**,**d**, none gave better results with respect to the parent stilbene derivatives **3c**, pterostilbene (**2a**), and resveratrol (**1a**) in terms of antiproliferative and proapoptotic activities. More interesting were the results obtained with the terphenyl derivatives **13a**−**i**. Among the compounds belonging to this class, the best results regarding the proapoptotic activity were obtained with the differently substituted derivatives **13e**, **13f**, and **13i**, suggesting that the ortho-substitution motif in the terphenyl architecture plays a prevalent role in determining this type of activity.

The lack of clearly identifiable SARs within the whole series as well as within each of the subsets (stilbenes, naphthalenes, terphenyls) points to a complex mechanism of action for all of these molecules. In principle, however, this is not surprising given their at least qualitative stereoelectronic similarity to resveratrol, whose mechanism of apoptosis induction is still controversial. We have already discussed the discrepancies of opinion about this aspect in a previous paper.[27]

Although the trihydroxylated terphenyl **13g** was not the most potent compound presently synthesized, it showed the most peculiar biological profile. In fact, **13g** caused a block of cell cycle in the G_0-G_1 phase, while resveratrol,[31a−e] as well as the other stilbenes or stilbene mimetics studied in this and previous works,[26,27] induced a recruitment of cells in S or G_2-M phases. Moreover, whereas at higher concentration **13g** was able to induce apoptosis both in sensitive and resistant cell lines, when used at low concentration (10 μM) for 72−96 h, it was able to induce a differentiation of HL60 cells. This is indeed a remarkable feature, if one considers that currently there is only one drug (*all-trans*-retinoic acid, ATRA) available in differentiation therapy to treat patient with acute promyelocytic leukemia (APL).[35a−b] Remarkably, however, the differentiating

effects of **13g** are, in part, different from those of ATRA, as the latter induces in vitro a prevalent granulocytic differentiation of APL blasts, while **13g** induces a granulocytic and monocytic differentiation.[36a−b] Moreover, no morphological aspects of macrophages able to phagocyte apoptotic cells have been described for ATRA.

Recently, Asou et al. first described the differentiating properties of resveratrol in a series of myeloblastic leukemia cells.[32] The differentiating effects of resveratrol were higher in NB4 and U935 cell lines but were poor in HL60 cells. The mechanism by which resveratrol induces differentiation is not known, although it has been suggested that a decrease in activity of NF-κB could be associated with the terminal differentiation induced by the compound. In the present work, we observed that resveratrol was able to induce the expression of differentiating markers in less than 25% of HL60 cells, while **13g** induced differentiation in about 60% of cells. Of interest, **13g** induced a myeloid differentiation of HL60 cells with the appearance of cells in different phases of differentiation; after 96 h of treatment, metamyelocytes and band neutrophils were the most representative cells. The presence of macrophages able to phagocyte apoptotic bodies suggests that differentiation induced by **13g** is not only morphological but also functional. Finally, in a previous work, some of us[37] observed that ATRA was unable to induce apoptosis in HL60 cells after 48 h of treatment; in contrast, **13g** was able to induce apoptosis after 24 h.

In conclusion, compared to resveratrol, the synthetic terphenyl **13g** showed a more potent apoptotic and differentiating activity. Moreover, **13g** was active on both MDR and Bcr-Abl-expressing cells that were resistant to resveratrol. For these characteristics, **13g** could be considered a promising lead to obtain proapoptotic and differentiating compounds for the treatment of acute leukemias. As a final observation from a chemical perspective, we want to remark that the terphenyl scaffold proved to be a versatile architecture for the identification of small molecules useful as chemical probes to study cell functions and mechanisms.

Experimental Section

General Parallel Procedure for the Synthesis of Derivatives 7a, b, 8−10. In five distinct reactors, 2-Br-naphthalenes **5** and **6** and *o*-, *m*-, and *p*-iodobromobenzene (1.0 equiv) were dissolved in toluene (10 mL). Then, 3,5-dimethoxyphenylboronic acid **4** (2 equiv) in EtOH (3 mL) and aqueous Na_2CO_3 (2 M, 3.0 equiv) were added to each reactor, and the resulting mixtures were deoxygenated with a stream of N_2. After 10 min, $Pd(PPh_3)_4$ (0.05 equiv) was added, and each mixture was brought to reflux, allowed to stir under N_2 for 5 h, cooled to room temperature, and treated as follows. Each solution was poured into a mixture of H_2O and Et_2O, and the two phases were separated. The aqueous layer was washed with Et_2O, and the organic phases were combined and washed with 1 M NaOH followed by brine. The ethereal solution was dried over Na_2SO_4 and evaporated. Purification of each crude product by flash chromathography using petroleum ether/ethyl acetate 9.5:0.5 or 9.9:0.1 yielded the corresponding (3,5-dimethoxyphenyl)naphthalenes **7a**,**b** and the biphenyldimethoxy derivatives **8−10**.

General Parallel Procedure for the Synthesis of Terphenyl Derivatives 13a−f. Each of the three bromobiphenyldimethoxy derivatives **8−10** (2.0 equiv) was dissolved in toluene (20 mL) and each was partitioned in two distinct reactors. 4-Substituted-phenylboronic acid **11** or **12** (2 equiv), in EtOH (3 mL), and aqueous Na_2CO_3 (2M, 3.0 equiv) were added to each set of three reactors containing **8−10**, and the resulting six mixtures were deoxygenated with a stream of N_2. After 10 min, $Pd(PPh_3)_4$ (0.05 equiv) was added, and each mixture was brought to reflux, allowed to stir under N_2 for 3−12 h, cooled to room temperature, and treated as follows. Each solution was poured into a mixture of H_2O and Et_2O, and the two phases were separated. The aqueous layer was washed with Et_2O, and the organic phases were combined and washed with 1 M NaOH followed by brine. The ethereal solution was dried over Na_2SO_4 and evaporated. Purification of each crude product by flash chromathography using petroleum ether/ethyl acetate yielded the corresponding terphenyl derivatives **13a−f**.

General Parallel Procedure for Demethylation to Trihydroxy Derivatives 7d, 13 g−i. In four distinct reactors, the trimethoxy derivatives **7a** and **13b,d,f** (1 equiv) were dissolved in anhydrous CH_2Cl_2 (10 mL) at −78 °C. Then, BBr_3 (1 M in CH_2Cl_2, 3 equiv) was added to each solution, and the resulting reaction mixture was allowed to warm to room temperature for 20 h, cooled at 0 °C, and treated as follows. Each solution was poured into H_2O, and the two phases were separated. The aqueous layer was washed twice with CH_2Cl_2, and the organic phases were combined and washed with a 1 M solution of sodium thiosulfate followed by H_2O. The organic layer was dried over Na_2SO_4 and evaporated to dryness. Purification of each crude product by flash chromathography using petroleum ether/ethyl acetate yielded the corresponding trihydroxy derivatives **7d**, **13g−i**.

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Supporting Information Available: General chemical methods, detailed biological protocols, general methods for the synthesis of stilbene derivatives **3a−h**, synthesis of 3,5-dimethoxyphenylboronic acid **4**, physical and spectroscopic data for compounds **3a−h**, **7a−d**, and **13a−i**, and elemental analysis results. This material is available free of charge via the Internet at http://pubs.acs.org.

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