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Synthesis and structure–activity relationships of new antiproliferative and proapoptotic retinoid-related biphenyl-4-yl-acrylic acids

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Abstract—Atypical retinoids, or retinoid-related molecules (RRMs), represent a class of proapoptotic agents with a promising potential in the treatment of neoplastic diseases. In the present work, the synthesis and structure–activity relationship of a series of 3′-adamantan-1-yl-biphenyl-4-yl-acrylic acids substituted in ring A were studied. The synthesized compounds were evaluated for their antiproliferative activity in a human promyelocitic leukemia cell line (NB4), and in an ovarian carcinoma cell system including IGROV-1, carrying a functional wild-type p53, and a cisplatin-resistant subline, IGROV-1/Pt-1. The presence of at least one oxygenated substituent in positions 4′ or 5′ appears determinant for the antiproliferative activity. With two substituents of this kind the activity increases, particularly in the case of alkylenedioxy compounds. The activation of DNA damage response as indicated by phosphorylation of H2AX histone, RPA-2 protein, and p53 at serine 15 by the most apoptotic compounds provides additional support to the hypothesis that the genotoxic stress is a critical event mediating apoptosis induction by compounds of this group.

1. Introduction

Retinoids may have clinical efficacy in cancer chemoprevention. The chemopreventive properties of agents of this class include growth inhibitory, prodifferentiating, proapoptotic, and antiangiogenic activities.[1,2] Synthetic retinoids containing the adamantyl moiety represent a promising group of novel agents of this class, because they are characterized by a potent proapoptotic activity in a large variety of tumor cells. They exhibit antitumor activity in in vivo models with acceptable side effects.[3] The detailed molecular mechanism involved in apoptosis induction by adamantyl retinoids is not clearly defined. These retinoids have been shown to induce apoptosis by a mechanism which is independent of retinoid receptor signaling pathway.[4–6]

We have recently reported that a novel adamantyl retinoid, ST1926 (**1**), is a potent inducer of apoptosis in a variety of tumor cell lines and exhibits efficacy against solid tumor models, with an improved pharmacological profile.[7] The evidence that ST1926 enhances the proapoptotic and antitumor activity of other clinically effective agents supports the potential interest of novel combination approaches with the use of agents which induce apoptosis through different mechanisms. The pattern of cellular response to ST1926 indicated the activation of DNA damage response, thus supporting that the induction of genotoxic stress is implicated in mediating drug-induced apoptosis.[8–10]

In our previous work we explored the putative key portions of the molecule, that is, the carboxylic group, the double bond, and the adamantan-1-yl moiety, by introducing substituents or structural modifications to these moieties.[7] We then evaluated the effects of the changes on the activity of the derivatives, using appropriate cellular and molecular models. The structure–activity relationship study suggested that the structural

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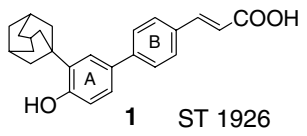

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Chart 1.

requirements for optimal activity of this class of compounds are rather strict, meaning that the carboxylic group, the *E* unsubstituted double bond, and a bulky lipophilic group in position 3′ are necessary to maintain high antiproliferative activity. Thus compound **1** remained the most potent compound of the series (Chart 1).

The present work was designed to gain further insight into the critical requirements linking structural features and biological activity of this class of biphenyl-4-yl-acrylic acids. Thus we planned to explore the effect of substituents on the ring A of the biphenyl system. In particular we wanted to evaluate the role of the oxygen atom in this portion of the ligand, first removing and then replacing the phenolic OH by other polar functions while maintaining the adamantyl group at the same position.

Interestingly, we noticed that removal of the phenolic OH group led to a strong decrease of cytotoxic activity, meaning that an oxygenated function is a critical structural feature. However, differently from other key portions of the molecule, where small changes in structure had a pronounced influence on biological activity, replacement of the phenolic OH or addition of other oxygenated groups in the adjacent position led to compounds endowed with potent antitumor activity, therefore worth of further investigation.

2. Chemistry

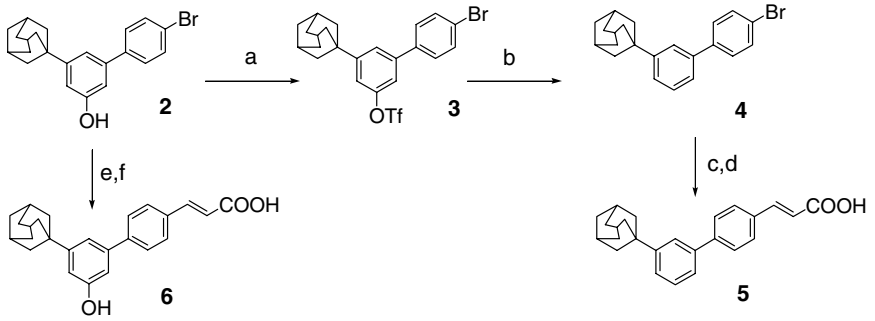
First, we planned to deoxygenate the phenol moiety of **1** to test the role of the hydroxy group. Attempts to obtain the desired compound from **1**, via the corresponding triflate, were unsuccessful. So we performed the reduction reaction starting from the triflate of the less hindered compound **2**, whose synthesis has been described in our previous paper.[7] We applied an efficient procedure that relies on the use of triethylsilane as a reducing agent and a catalytic system consisting of palladium(II)acetate and the bidentate phosphine ligand 1,3-bis(diphenylphosphino)propane (dppp). The reaction was highly chemoselective and the bromo group was completely unaffected under these conditions.[11]

The synthesis of compound **6**, in which the OH group was moved from its original position, has already been described (Scheme 1).[7]

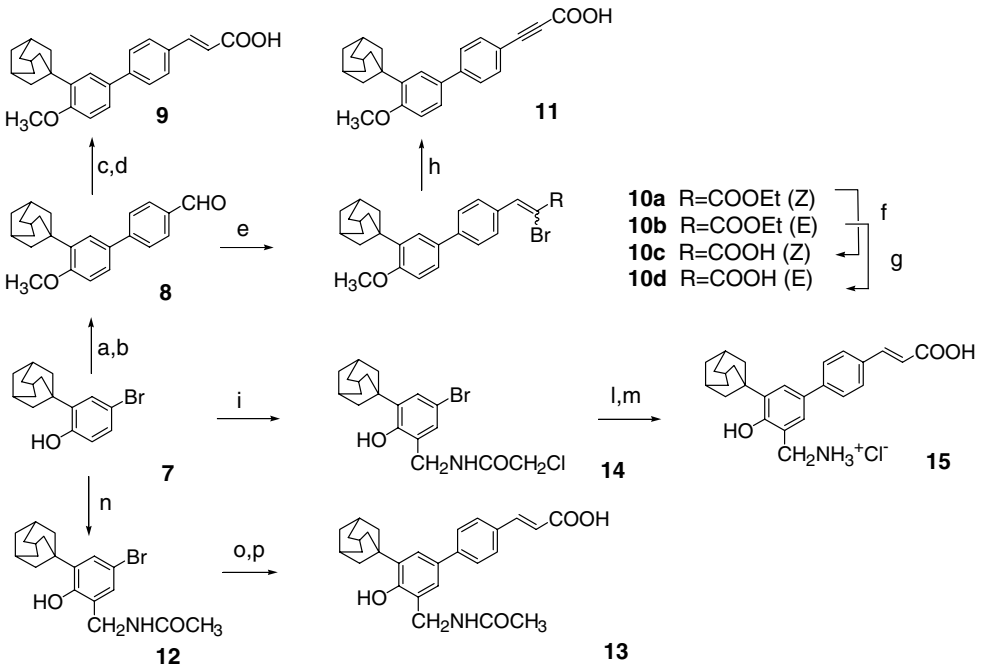
Compounds **9**–**15** were obtained as outlined in Scheme 2. First, 2-(1-adamantyl)-4-bromophenol[7] **7** was alkylated with methyl iodide and then cross-coupled with *p*-formylbenzeneboronic acid to give the aldehyde **8**. Wittig condensation with methoxycarbonylmethylenetriphenylphosphorane and final basic hydrolysis led to compound **9**. Condensation of aldehyde **8** with carbethoxybromomethylenetriphenylphosphorane led to a mixture of the two diastereomeric esters **10a** and **10b**, that were separated by chromatography. Basic hydrolysis of **10b** led to the corresponding acid **10d**, while hydrolysis of *Z*-**10a** afforded the elimination product **11** together with the acid **10c**.

The introduction of an aminomethyl moiety was accomplished by alkylation of compound **7**[7] followed by palladium-catalyzed cross-coupling with methyl *p*-bromocinnamate and hydrolysis with HCl 37% in dioxane to give the acid **15**. The corresponding acetyl derivative **13** was obtained by Friedel–Crafts (FC) alkylation of **7** with N-(hydroxymethyl)-acetamide, Suzuki coupling, and basic hydrolysis.

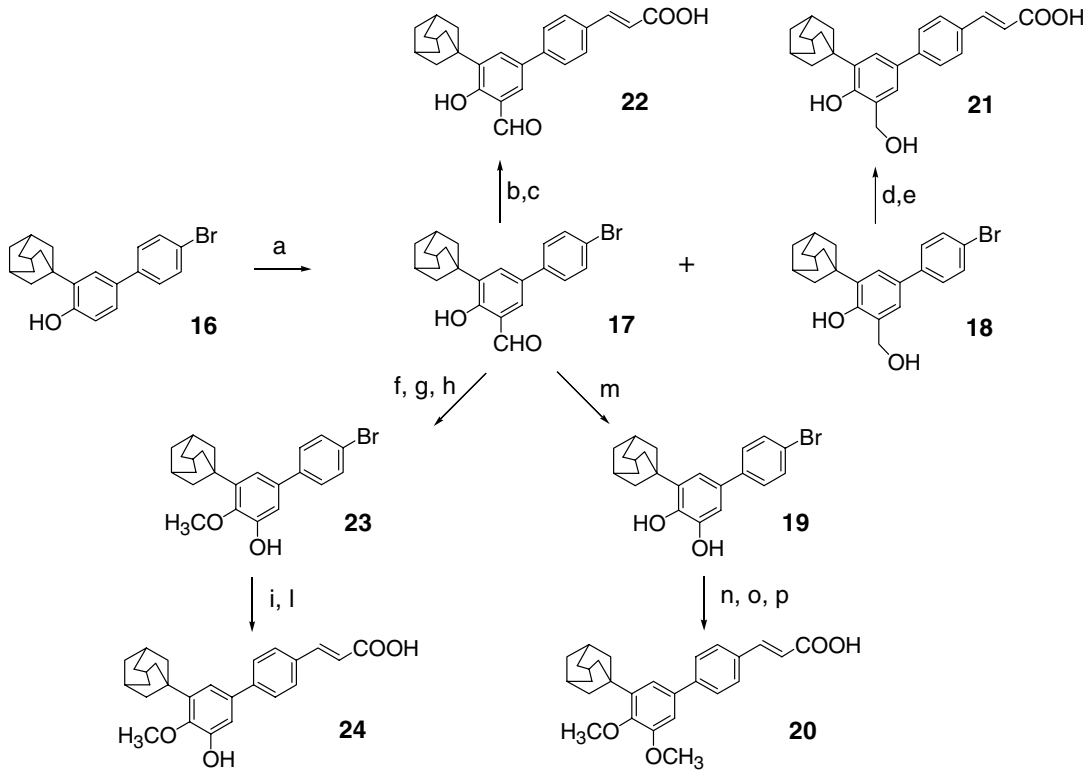
Compounds substituted with two oxygenated moieties were prepared as depicted in Schemes 3–5. A FC alkylation of 4′-bromobiphenyl-4-ol with adamantan-1-ol led to compound **16**. Formylation of this latter by $(CH_2O)_n$[12] led mainly to compound **17**, that was converted into the acid **22** by a palladium-catalyzed cross-coupling Heck reaction followed by hydrolysis. Compound **18**, obtained in small amount from the formylation reaction, was used as a starting material for the synthesis of **21**, with the usual sequence of reactions.



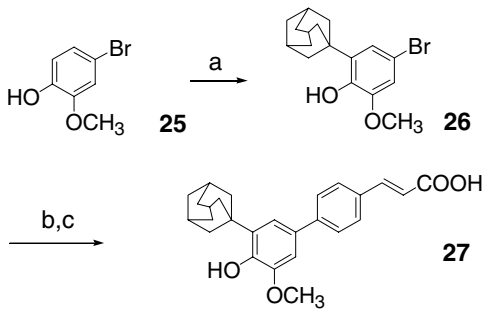
Scheme 1. Reagents and conditions: (a) TfO_2, Py, 3 h, rt, 87%; (b) Et_3 SiH/dppp, $Pd(OAc)_2$, DMF, 24 h, 60 °C, 42%; (c) TOTP, $Pd(OAc)_2$, TEA, methyl acrylate, 32 h, reflux, 67%; (d) LiOH·H_2O, THF/H_2O, rt, overnight, 63%; (e) TOTP, $Pd(OAc)_2$, TEA, methyl acrylate, 2 days, reflux, 50%; (f) LiOH·H_2O, THF/H_2O, rt, overnight, 80%.



Scheme 2. Reagents and conditions: (a) NaH, MeI, DMF, 2 h, 20 °C 78%; (b) Pd tetrakis, 4-formylbenzeneboronic acid, 2 M Na_2CO_3, toluene, 2 h, reflux, 72%; (c) Ph_3 PCH = COOMe, chloroform, 3 h, reflux, 86%; (d) NaOH, methanol, 7 h, reflux, 70%; (e) Ph_3PCBr = COOEt, chloroform, 14 h, reflux, 87%; (f) KOH, methanol, 1 h, reflux, 7%; (g) LiOH, THF/H_2O, rt, overnight, 89%; (h) KOH, methanol, 1 h, reflux, 68%; (i) $ClCH_2CONHCH_2OH$, CH_3COOH/H_2SO_4 9:1, 20 h, rt, 87%; (l) bis(pinacolato)diboron, KOAc, $PdCl_2$(dppf), then methyl *p*-Br-cinnamate, $PdCl_2$(dppf), Na_2CO_3 2 M, 3 h, 100 °C, 27% ; (m) HCl 37%, dioxane, 23 h, reflux, 92%; (n) $CH_3CONHCH_2OH$, $CH_3COOH:H_2SO_4$ 9:1, 5 days, rt, 37%; (o) bis(pinacolato)diboron, KOAc, $PdCl_2$(dppf), then methyl *p*-Br-cinnamate, $PdCl_2$(dppf), Na_2CO_3 2 M, 100 °C, 3 h, 43% ; (p) LiOH THF/H_2O, rt, overnight, 72%.



Scheme 3. Reagents and conditions: (a) $(CH_2O)n$, $SnCl_4$, 2,6-lutidine, toluene, 7 h, 95 °C, 60% (17), 34% (18); (b) methyl acrylate, TOTP, Et_3N, $Pd(OAc)_2$, 110 °C, 66%; (c) LiOH THF/H_2O, rt, overnight, 83%; (d) methyl acrylate, TOTP, Et_3N, $Pd(OAc)_2$, 110 °C, 33%; (e) LiOH THF/H_2O, rt, overnight, 91%; (f) CH_3I, K_2CO_3, acetone, 6 h, reflux, 90%; (g) MCPBA, CH_2Cl_2, 1 h, reflux, 47%; (h) KOH, MeOH, 1 h, rt, then HCl, 100%; (i) methyl acrylate, TOTP, Et_3N, $Pd(OAc)_2$, 67%; (l) NaOH, methanol, 2 h, reflux, 20% (m) MCPBA, CH_2Cl_2, 20 h, reflux, 53%; (n) methyl acrylate, TOTP, Et_3N, $Pd(OAc)_2$, 84%; (o) CH_3 I, NaH, DMF, rt, 2 h, 19%; (p) LiOH THF/H_2O, rt, 4 days, 87%.



Scheme 4. Reagents and conditions: (a) 1-adamantanol, CH_2Cl_2, concd H_2SO_4, rt, 1 h, 60%; (b) bis(pinacolato)diboron, $PdCl_2$(dppf), KOAc, methyl *p*-bromocinnamate, Na_2CO_3 2 M and $PdCl_2$(dppf), 70%; (c) LiOH THF/H_2O, rt, overnight, 98%.

Methylation of **17** with CH_3I in acetone, followed by Baeyer–Villiger oxidation[13] and hydrolysis with KOH in methanol, afforded compound **23**. The acid **24** was then obtained by Heck coupling and hydrolysis. Baeyer–Villiger oxidation on the aldehyde **17** gave the catechol **19** that was immediately subjected to Heck coupling with methyl acrylate. The high instability of the obtained catechol required methylation of both the phenolic groups with methyl iodide and NaH in DMF. Hydrolysis of the ester gave the desired carboxylic acid **20**.

The synthesis of compounds **27**–**35** is illustrated in Schemes 4,5. Friedel–Crafts alkylation of 2-methoxy-4-bromophenol led to compound **26**. Palladium-catalyzed Suzuki one-pot cross-coupling with bis(pinacolato)diboron and methyl *p*-bromocinnamate[14] followed by the usual saponification with LiOH furnished the acid **27**.

Aldehyde **30** was obtained by FC alkylation and palladium-catalyzed cross-coupling of the bromobenzene **29a**. Wittig condensation with methoxycarbonylmethylenetriphenylphosphorane followed by standard base hydrolysis conditions led to acid **31a**. The acid **31b** was prepared via palladium-catalyzed Suzuki one-pot condensation with methyl 4-bromocinnamate followed by hydrolysis.

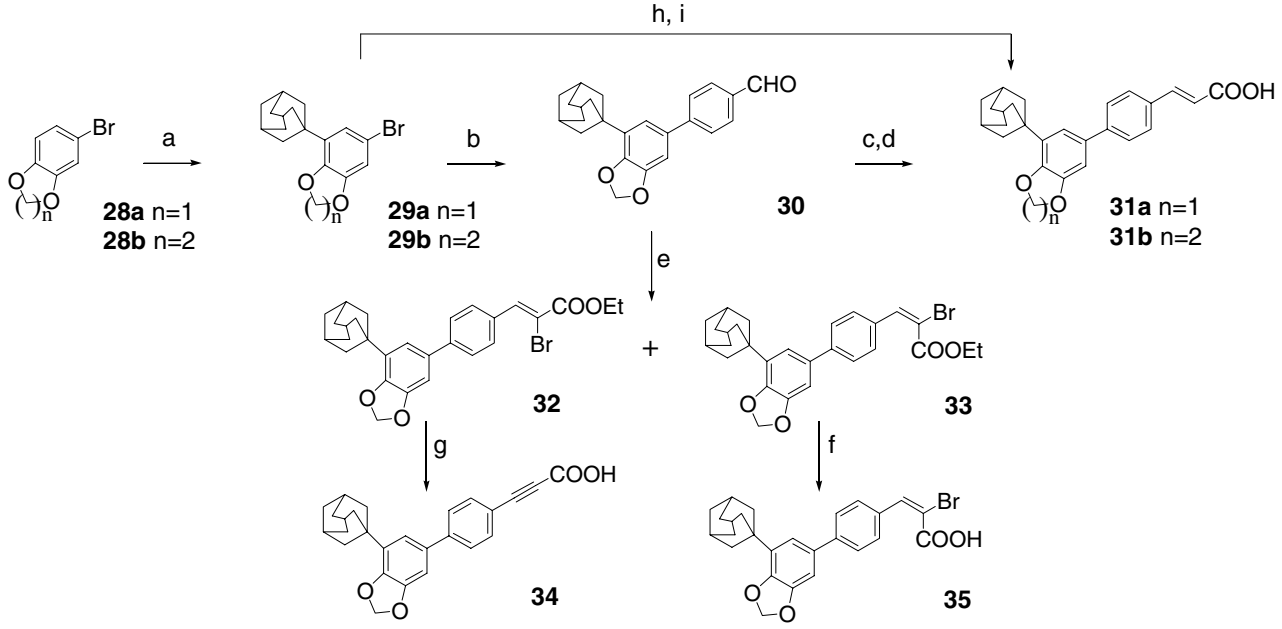
The two diastereomeric esters **32** and **33** were obtained by Wittig condensation of **30a** with ethoxycarbonylbromomethyltriphenylphosphorane. Final hydrolysis, accompanied by elimination in the case of **32**, led to acids **34** and **35**.

3. Antiproliferative activity

The effects of the selected compounds on cell growth were determined in a panel of human tumor cells, including an ovarian carcinoma cell line (IGROV-1), a subline selected for resistance to cisplatin and characterized by p53 mutation (IGROV-1/Pt1), and a leukemia cell line (NB4). The results are reported in Table 1.

4. Cytodifferentiation test

The obtained compounds were also tested in vitro for their cytodifferentiating activity on the leukemia cell line NB4.



Scheme 5. Reagents and conditions: (a) 1-adamantanol, CH_2Cl_2, concd H_2SO_4, 4 h, rt; (b) $Pd(PPh_3)_4$, 4-formylbenzeneboronic acid, Na_2CO_3 2 M, EtOH, toluene, 7 h, reflux, 70%; (c) Ph_3P = CHCOOMe, $CHCl_3$; (d) LiOH, THF/H_2O; (e) Ph_3P = CBrCOOEt, $CHCl_3$, 8 h, reflux; (f) LiOH, THF/H_2O 89%; (g) KOH, MeOH, 1 h, reflux, 40%; (h) bis(pinacolato)diboron, KOAc, $PdCl_2$(dppf), methyl *p*-Br-cinnamate, Na_2CO_3, 50%; (i) LiOH, THF/H_2O, rt, overnight, 91%.

Table 1. Antiproliferative activity of selected compounds on IGROV-1, IGROV-1/Pt1, and NB4 cellular lines

Compound	IC_{50} (μM)		
	IGROV-1	IGROV-1/Pt1	NB4
1	0.23 ± 0.08	0.40 ± 0.08	0.082 ± 0.005
5	18.8	21.9	n.d.
6	1.31 ± 0.02	n.d.	1.2 ± 0.09
9	1.19 ± 0.5	1.43 ± 0.58	1.1 ± 0.07
10d	30.6	>10	41 ± 2.4
10c	≫20	≫20	17.9 ± 2.2
11	1.70 ± 0.60	4.8 ± 1.9	n.d.
13	1.24 ± 0.07	2.89 ± 0.02	0.99 ± 0.03
15	0.45 ± 0.3	0.39 ± 0.18	0.18 ± 0.008
20	0.71 ± 0.10	2.4 ± 0.57	1.89 ± 0.07
21	0.76 ± 0.57	1.04 ± 0.57	0.19 ± 0.01
22	1.64 ± 0.03	2.72 ± 0.57	1.61 ± 0.1
24	0.57 ± 0.14	0.97 ± 0.33	0.55 ± 0.03
27	0.52	n.d.	0.27 ± 0.02
31a	0.39 ± 0.17	0.46 ± 0.09	0.26 ± 0.009
31b	0.23 ± 0.07	0.48 ± 0.25	0.11 ± 3.4
34	3.6	4.80 ± 2.4	2.0 ± 0.03
35	36.6	34.10	72.3 ± 4.4

IC_{50} (μM) is the concentration required for 50% inhibition of cell growth (±SD).

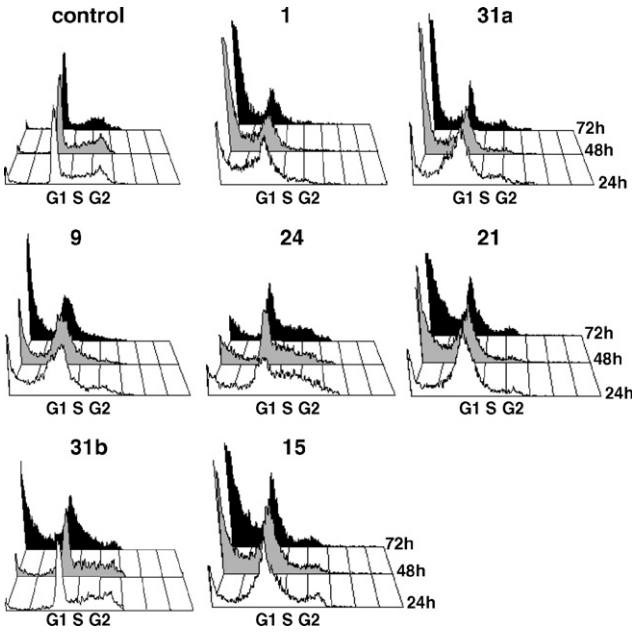


Figure 1. Time course of cell cycle perturbations in ovarian carcinoma IGROV-1 cells, following treatment with selected retinoids. Cells were treated with the drug at IC_{80} and the cell cycle was analyzed by FACScan analysis of PI-stained cells at 24, 48, and 72 h of treatment. One experiment representative of three is reported.

5. Results and discussion

As a first step we checked the role of the phenolic OH group in 4′ position, by preparing the deoxy compound **5**. This modification induced a dramatic decrease of the antiproliferative activity, thus indicating the importance of such a group for the activity. Shifting the OH to the 5′ position (compound **6**) or methylation of the 4′-OH (compound **9**) restored the activity, so that the minimum requirement is the presence of an oxygen atom in at least one of the two positions. The introduction of two oxygenated functions (compounds **20**, **24**, **27**, **31a**, **31b**) substantially maintained the activity, the two compounds with a ring (**31a**, **31b**) being the most active. Probably in this case the oxygen atoms are more exposed to the interaction with the receptor with respect to the less accessible oxygens in the methoxy derivatives. Also the introduction of a carbon atom linked to a heteroatom in the position adjacent to the OH (compounds **13, 15, 21, 22**) maintained the activity.

For two compounds (**9** and **31a**) some modifications in the distal part of the molecule were made, in order to check whether the structural requirements for activity held also in this series.[7] In fact the compounds with a bromoacrylic chain **10c–d** and **35** appeared inactive, whereas the propynoic acid derivatives (**11** and **34**) maintained the activity, as expected.[7]

In this new series compound **31b** was the most potent agent with activity comparable with **1** in all tested cell lines. Cell cycle analysis of drug-treated cells revealed a marked perturbation of cell cycle progression (Fig. 1). The most active compounds of this series (e.g., **9**, **24**, **31a**, **31b**, **15**) caused accumulation of cells in the G1/S phases with the appearance in a time-dependent manner of a sub-G1 peak, which is considered to be a marker of cell death. This behavior was less marked for compound **24**.

All compounds effective in cell growth inhibition were also active as proapoptotic agents (Table 2). Compound **24** exhibited an appreciably reduced ability to induce apoptosis. This observation was consistent with a lower sub-G1 peak (Fig. 1). All tested compounds were able to activate DNA damage response as indicated by phosphorylation of H2AX histone, RPA-2 protein, and p53 at serine 15 (Fig. 2). Compound **9** was the most effective in inducing phosphorylation of H2AX histone, which is known to be the most sensitive marker of double-strand DNA breaks.[15] Again compound **24**, which is a weaker apoptosis inducer, was also less effective in activation of a genotoxic stress response. This observation provides additional support to the hypothesis that the genotoxic stress is a critical event mediating apoptosis induction by compounds of this groups.

Table 2. Apoptosis induction in IGROV-1 cells by selected retinoids

IGROV-1	% of apoptosis
1	51 ± 3
15	55
31a	50 ± 5
9	47 ± 5
21	45
31b	30 ± 4
24	20 ± 4

Apoptosis was determined at 72 h following exposure to IC_{80} by morphological analysis of propidium iodide-stained cells. The results are expressed as percentage of apoptotic cells versus total cell number. Values are the mean standard deviation of three independent experiments.

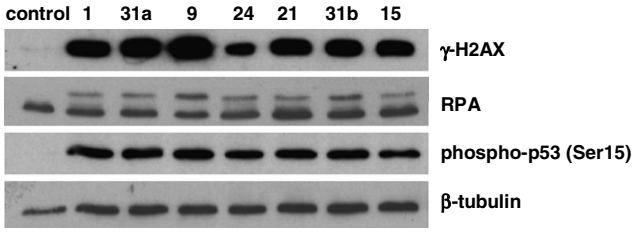


Figure 2. Induction of DNA damage response in IGROV-1 cells exposed to equitoxic drug concentration of selected retinoids (IC_{80}) for 6 h. Cells were lysed and processed for Western blotting. Control loading is shown by β-tubulin.

As already reported for compound **1**,[8] atypical adamantyl retinoids caused modulation of MAP kinases implicated in stress response (e.g., p38 and JNK). We investigated the ability of **31a** to activate these kinases as detected by their enhanced phosphorylation (Fig. 3). In IGROV-1 cells, activation of p38 and JNK was more marked following exposure to **31a**, in spite of a comparable proapoptotic activity. Activation of stress kinases by compound **1** was more evident in IGROV-1/Pt1 cells, characterized by p53 mutation and by reduced sensitivity to these agents, thus indicating no correlation between stress response and apoptosis. These observations suggest that the activation of MAP kinase pathways plays a protective role in the cellular stress response to these agents.

A preliminary evaluation of the ability of compounds of this series to induce differentiation of NBA leukemia cells indicated that some compounds had cytodifferentiating activity comparable to (**10c**, **11**) or better (**9**, **31b**) than the standard ATRA (all-*trans* retinoic acid) in a dose-dependent manner, thus supporting an additional therapeutic interest for these compounds.

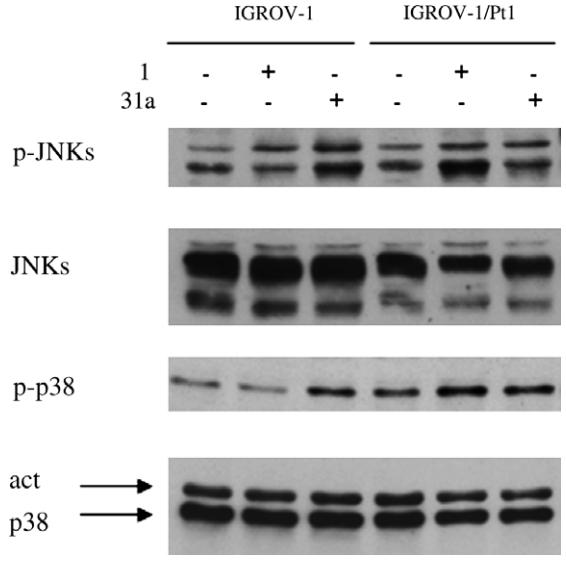


Figure 3. Effect of compounds **1** and **31a** on JNK and p38 activation in IGROV-1 and IGROV-1/Pt1 cells exposed to the drugs at their respective IC_{80}. Western blot analysis was performed with phospho-specific antibodies after 18 h of treatment. Anti-p38 and JNK blots are shown as control of protein expression and anti-actin as control of protein loading.

In conclusion, we have explored which structural features confer strong antiproliferative activity and proapoptotic properties in a class of atypical retinoids. These results can be useful for the modulation of the pharmacological potential of these compounds.

6. Experimental

6.1. General chemical methods

All reagents and solvents were of reagent grade or were purified by standard methods before use. Melting points were determined in open capillaries on a Büchi melting point apparatus and are uncorrected. Column chromatography was carried out on flash silica gel (Merck 230–400 mesh). TLC analysis was conducted on silica gel plates (Merck $60F_{254}$). NMR spectra were recorded at 300 MHz with a Bruker instrument. Chemical shifts (δ values) and coupling constants (J values) are given in ppm and Hz, respectively. Mass spectra were recorded at an ionizing voltage of 70 eV on a Finnigan TQ70 spectrometer. The relative intensities of mass spectrum peaks are listed in parentheses. HPLC analysis of the mixture of diastereoisomers was performed on an HP 1050 quaternary pump fitted with a Rheodyne injector (20 μL loop) and a HP-1050 Diode-Array detector. Chromatograms were recorded at 360 and 400 nm. The column was a Rainin C18, 25×0.4 cm, flow 1 mL/min, with a gradient from CH_3CN/H_2O 30:70 to 100:0, in 20 min.

6.2. General procedure for Heck condensation (A)

A round flask containing a mixture of 0.73 mmol of the appropriate aryl bromide, 1.17 mmol of methyl acrylate, 5 mg of Pd $(OAc)_2$, and 27 mg of tri-*o*-tolylphosphine in 0.3 mL of Et_3N was immersed in an oil bath preheated at 50 °C and kept at 100–110 °C for 2–12 h. Iced water was then added, followed by 2 N HCl. The aqueous phase was extracted repeatedly with ethyl acetate, then the organic phases were dried over Na_2SO_4, filtered, and evaporated. The crude product was purified by flash chromatography and, when necessary, crystallized from diisopropyl ether.

6.3. General procedure for ester hydrolysis (B)

The appropriate ester (0.16 mmol) was suspended in a solution of 34 mg (0.815 mmol) of $LiOH{\cdot}H_2O$ in 8.3 mL of THF/H_2O 3:2 and stirred at room temperature in the dark overnight. After evaporation of THF the remaining aqueous phase was washed with hexane, diethyl ether and then acidified with HCl 2 N (0.4 mL). The precipitated white solid was filtered and dried. No further purification was necessary.

6.4. General procedure for one-pot Suzuki condensation (C)

A small amount of the appropriate aryl bromide (0.6 mmol), 166 mg (0.65 mmol) of bis(pinacolato)diboron, 174 mg (1.78 mmol) of KOAc, and 13 mg

(0.018 mmol) of $PdCl_2$(dppf) were dissolved in 36 mL of dioxane and the mixture was stirred at 100 °C for 1–2 h under nitrogen. After cooling the solution at room temperature, methyl 4-bromocinnamate (287 mg, 1.19 mmol), $PdCl_2$(dppf) (13 mg, 0.018 mmol) and 2 M Na_2CO_3 (1.48 mmol, 0.74 mL) were added and the mixture was heated at 80–100 °C for 1–3 h. The solution was cooled at room temperature, diluted with water, acidified with 2 N HCl (2.4 mL), and extracted with ethyl acetate. The organic phase was washed with brine, dried over Na_2SO_4, filtered, and evaporated. The crude product was purified by flash chromatography.

6.4.1. Trifluoromethanesulfonic acid 4′-bromo-5-(adamantan-1-yl)-biphenyl-3-yl ester (3). To a solution of 5-(1-adamantanyl)-4′-bromobiphenyl-3-ol (**2**)[7] (840 mg, 2.19 mmol) in 10 mL of pyridine at 0 °C trifluoromethanesulfonic anhydride (0.41 mL, 2.41 mmol) was slowly added. The resulting mixture was stirred at 0 °C for 30 min and then allowed to warm at room temperature and stirred for 3 h. The mixture was then poured into water and extracted with diethyl ether. The organic extracts were washed sequentially with water, 2 N HCl, water, brine, dried, and concentrated to give a yellow oil. Purification by flash chromatography (hexane/ethyl acetate 95:5) afforded 980 mg (87%) of the title compound. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.73 (6H, s, 6Ad), 1.95 (3H, s, 3Ad), 2.10 (6H, s, 6Ad), 7.20 (2H, br s, 2Ar), 7.35 (2H, m, 2Ar), 7.50 (1H, s, 1Ar), 7.58 (2H, m, 2Ar). Anal. calcd for $C_{23}H_{22}BrF_3O_3S$: C, 53.60; H, 4.30. Found: C, 53.75; H, 4.18.

6.4.2. 4′-Bromo-3-(adamantan-1-yl)-biphenyl (4). To a mixture of the above triflate (68 mg, 0.13 mmol), $Pd(OAc)_2$ (0.6 mg, 2.6.10–3 mmol), and dppp (1.1 mg 2.6.10–3 mmol) in 1 mL of DMF at 60 °C was added Et_3SiH (0.325 mmol, 52.5 μL). The stirring was continued for 1.5 h and during this time the solution changed color from red to yellow. After dilution with diethyl ether the mixture was successively washed with water, saturated aqueous sodium bicarbonate, and brine, then it was dried and evaporated. The crude was purified by flash chromatography (hexane/dichloromethane 99:1) to obtain 20 mg (42%) of the title compound. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.85 (6H, s, 6Ad), 2.01 (3H, s, 3Ad), 2.15 (6H, s, 6Ad), 7.30–7.60 (8H, m, 8Ar). Anal. calcd for $C_{22}H_{23}Br$: C, 71.94; H, 6.31. Found: C, 71.86; H, 6.35.

6.4.3. 3-(3′-(Adamantan-1-yl)-biphenyl-4-yl)-acrylic acid (5). Compound **4** (100 mg, 0.27 mmol) was reacted with methyl acrylate according to the general procedure A, to obtain 67 mg (67%) of 3-(3′-(1-adamantyl)-biphenyl-4-yl)-acrylic acid methyl ester, mp 122–123 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.73 (6H, s, 6Ad), 1.95 (3H, s, 3Ad), 2.10 (6H, s, 6Ad), 3.78 (3H, s, OMe), 6.48 (1H, d, CH=, J = 16.0 Hz), 7.35–7.40 (3H, m, 3Ar), 7.50–7.65 (5H, m, 5Ar), 7.75 (1H, d, CH=, J = 16.0 Hz). The above ester (67 mg, 0.18 mmol) was hydrolyzed according to the general procedure B, to afford 40 mg (63%) of the title compound, mp 268–269 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.67 (6H, s, 6Ad), 1.98 (3H, s, 3Ad), 2.03 (6H, s, 6Ad), 6.55 (1H, d, CH=, J = 16.0 Hz), 7.30–7.80 (9H, m, 8Ar + CH=). Anal. calcd for $C_{25}H_{26}O_2$: C, 83.76; H, 7.31. Found: C, 83.84; H, 7.22.

6.4.4. 4′-Methoxy-3′-(adamantan-1-yl)biphenyl-4-carbaldehyde (8). To a suspension of sodium hydride (60% in mineral oil, 0.402 g, 10.1 mmol) in 11 mL of DMF, 2-(1-adamantyl)-4-bromophenol (**7**) (2.75 g, 8.38 mmol) was slowly added, while maintaining the temperature at 20 °C. The mixture was stirred at room temperature for 1 h, then methyl iodide (0.73 mL, 11.7 mmol) was added. After having stirred for additional 2 h at 20 °C, the solution was poured into water and extracted with diethyl ether. The organic phase was dried and evaporated. The residue was purified by flash chromatography (hexane) to give 2.1 g (78%) of 4-bromo-1-methoxy-2-(adamantan-1-yl)-benzene, mp 136 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.75 (6H, s, 6Ad), 2.05 (9H, s, 9Ad), 3.80 (3H, s, OMe), 6.72 (1H, d, 1Ar, J = 8.0 Hz), 7.24 (1H, dd, 1Ar, J = 8.0, 2.2 Hz), 7.28 (1H, d, 1Ar, J = 2.2 Hz).

To a solution of 1.95 g (6.06 mmol) of 4-bromo-1-methoxy-2-(adamantan-1-yl)-benzene in 12.2 mL of toluene, 6.06 mL (12.1 mmol) of 2 M aqueous Na_2CO_3, 0.210 g (0.182 mmol) of tetrakis-triphenylphosphine-palladium and a solution of 1 g (6.67 mmol) of 4-formylbenzeneboronic acid in 2.83 mL of ethanol were added. The mixture was refluxed for 2 h under a stream of nitrogen, then it was cooled at room temperature, diluted with dichloromethane, and washed with brine. The organic phase was dried, filtered, and evaporated. The residue was purified by flash chromatography (hexane/ethyl acetate 85:15) to give 1.5 g (72%) of the desired aldehyde, mp 196 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.80 (6H, s, 6Ad), 2.10 (9H, s, 9Ad), 3.90 (3H, s, OMe), 6.98 (1H, d, 1Ar, J = 8.0 Hz), 7.47 (1H, dd, 1Ar, J = 8.0, 2.2 Hz), 7.48 (1H, d, J = 2.2 Hz), 7.70 (2H, d, 2Ar, J = 8.3 Hz), 7.90 (2H, d, 2Ar, J = 8.3 Hz), 10.0 (1H, s). Anal. calcd for $C_{24}H_{26}O_2$: C, 83.20; H, 7.56. Found: C, 83.08; H, 7.70.

6.4.5. 3-(4′-Methoxy-3′-(adamantan-1-yl)biphenyl-4-yl)-acrylic acid (9). Compound **8** (658 mg, 1.90 mmol) and 635 mg (1.90 mmol) of methoxycarbonylmethylenetriphenylphosphorane were dissolved in 10 mL of chloroform and the solution was refluxed for 3 h in a current of nitrogen. Chloroform was evaporated and the crude product was purified by flash chromatography (dichloromethane/hexane 40:60) to give 655 mg (86%) of 3-(4′-methoxy-3′-(adamantan-1-yl)biphenyl-4-yl)-acrylic acid methyl ester, mp 212 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.73 (6H, s, 6Ad), 2.04 (3H, s, 3Ad), 2.10 (6H, s, 6Ad), 3.76 (3H, s, OMe), 3.82 (3H, s, OMe), 6.40(1H, d, CH=, J = 17,0 Hz), 6.88 (1H, d, 1Ar, J = 8.6 Hz), 7.35 (1H, dd, 1Ar, J = 8.6, 2.2 Hz), 7.41 (1H, d, 1Ar, J = 2.2 Hz), 7.45–7.60 (4H, m, 4Ar), 7.67 (1H, d, CH=, J = 17,0 Hz). The above ester (120 mg, 0.298 mmol) was suspended in 17 mL of a solution of 0.7 N NaOH in methanol and the mixture was refluxed for 7 h. After evaporation of methanol, the residue was treated with water, acidified with 6 N HCl, and the precipitate was filtered to afford 80 mg (70%) of the pure

product, mp >240 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.67 (6H, s, 6Ad), 1.98 (3H, s, 3Ad), 2.03 (6H, s, 6Ad), 3.74 (3H, s, OMe), 6.45 (1H, d, CH═, J = 16.2 Hz), 6.97 (1H, d, 1Ar, J = 8.5 Hz), 7.34 (1H, d, 1Ar, J = 2.3 Hz), 7.43 (1H, dd, 1Ar, J = 8.5, 2.3 Hz), 7.53 (1H, d, CH═, J = 16.2 Hz), 7.57 (2H, d, 2Ar, J = 8.4 Hz), 7.65 (2H, d, 2Ar, J = 8.4 Hz). Anal. calcd for $C_{26}H_{28}O_3$: C, 80.38; H, 7.26. Found: C, 80.34; H, 7.31.

6.4.6. *Z*-2-Bromo-3-[4′-methoxy-3′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid ethyl ester (10a) and *E*-2-bromo-3-[4′-methoxy-3′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid ethyl ester (10b). A solution of 2.78 g (8.02 mmol) of aldehyde **8** and 3.43 g (8.02 mmol) of carbethoxybromomethylenetriphenylphosphorane in 43 mL of chloroform was refluxed for 14 h under nitrogen. The solvent was evaporated and the residue was purified by flash chromatography (hexane/dichloromethane 45:55) to obtain 2.89 g of the *Z* diastereoisomer (**10a**), and 700 mg of the *E* diastereoisomer (**10b**). Yield 87%, mp (Z) 129 °C, mp (E) 135 °C. ^{1}H NMR *Z* isomer (300 MHz, $CDCl_3$) δ: 1.42 (3H, t, Et, J = 6.0 Hz), 1.83 (6H, s, 6Ad), 2.14 (9H, s, 9Ad), 3.90 (3H, s, OMe), 4.37 (2H, q, Et, J = 6.0 Hz), 6.97 (1H, d, 1Ar, J = 8.0 Hz), 7.45 (1H, dd, 1Ar, J = 8.0, 2.2 Hz), 7.54 (1H, d, 1Ar, J = 2.2 Hz), 7.65 (2H, d, 2Ar, J = 8.4 Hz), 7.95 (2H, d, 2Ar, J = 8.4 Hz), 8.25 (1H, s, CH═). Anal. calcd for $C_{28}H_{31}BrO_3$: C, 67.88; H, 6.31. Found: C, 67.73; H 6.39.

^{1}H NMR *E* isomer (300 MHz, $CDCl_3$) δ: 1.42 (3H, t, Et, J = 5.0 Hz), 1.80 (6H, s, 6Ad), 2.10 (3H, s, 3Ad), 2.15 (6H, s, 6Ad), 3.87 (3H, s, OMe), 4.25 (2H, q, Et, J = 5.0 Hz), 6.93 (1H, d, 1Ar, J = 8.3 Hz), 7.33 (2H, d, 2Ar, J = 8.5 Hz), 7.37 (1H, s, CH═), 7.40 (1H, dd, 1Ar, J = 8.3, 2.2 Hz), 7.48 (1H, d, 1Ar, J = 2.2 Hz), 7.52 (2H, d, 2Ar, J = 8.5 Hz). Anal. calcd for $C_{28}H_{31}BrO_3$: C, 67.88; H. 6.31. Found: 67.71; H, 6.34.

6.4.7. *E*-3-(3′-Adamantan-1-yl-4′-methoxy-biphenyl-4-yl)-2-bromo-acrylic acid (10d). The ester **10b** (150 mg, 0.303 mmol) was hydrolyzed according to the general procedure B for one night in the dark. The precipitated solid was filtered to give 126 mg (89%) of the pure product, mp 234 °C, ^{1}H NMR (300 MHz, DMSO/d_6) δ: 1.70 (6H, s, 6Ad), 2.00 (3H, s, 3Ad), 2.08 (6H, s, 6Ad), 3.80 (3H, s, OMe), 7.05 (1H, d, 1Ar, J = 8.5 Hz), 7.35 (1H, s, CH═), 7.40 (1H, d, 1Ar, J = 2.4 Hz), 7.42 (2H, d, 2Ar, J = 8.4 Hz), 7.50 (1H, dd, 1Ar, J = 8.5, 2.4 Hz), 7.65 (2H, d, 2Ar, J = 8.4 Hz). Anal. calcd for $C_{26}H_{27}BrO_3$: C, 66.81; H, 5.82. Found: C, 66.90; H, 5.87.

6.4.8. [4′-Methoxy-3′-(adamantan-1-yl)biphenyl-4-yl]propynoic acid (11) and *Z*-3-(3′-(Adamantan-1-yl)-4′-methoxy-biphenyl-4-yl)-2-bromo-acrylic acid (10c). A suspension of 2.89 g (5.85 mmol) of the ester **10a** in 16 mL of a 25% solution of potassium hydroxide in methanol was refluxed for 1 h. The solvent was evaporated and water was added to the residue, then HCl 12 N was slowly added. The formed solid was filtered and dried. Crystallization from diisopropylether gave 1.5 g (68%) of compound **11**, mp 246 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.75 (6H, s, 6Ad), 2.09 (9H, s, 9Ad), 3.85 (3H, s, OMe), 7.08 (1H, d, 1Ar, J = 8.0 Hz), 7.44 (1H, d, 1Ar, J = 2.0 Hz), 7.55 (1H, dd, 1Ar, J = 8.0, 2.0 Hz), 7.65 (2H, d, 2Ar, J = 8.0 Hz), 7.72 (2H, d, 2Ar, J = 8.0 Hz). MS (EI) *m*/*z* 342 (M−CO_2). A small amount (7%) of acid **10c** was obtained as a side product of this reaction. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.70 (6H, s, 6Ad), 2.05 (3H, s, 3Ad), 2.10 (6H, s, 6Ad), 3.85 (3H, s, OMe), 7.05 (1H, d, 1Ar, J = 8.3 Hz), 7.43 (1H, d, 1Ar, J = 1.2 Hz), 7.53 (1H, dd, 1Ar, J = 8.3, 1.2 Hz), 7.70 (2H, d, 2Ar, J = 8.3 Hz), 7.96 (2H, d, 2Ar, J = 8.3 Hz), 8.25 (1H, s, CH═). Anal. calcd for $C_{26}H_{27}BrO_3$: C, 66.81; H, 5.82. Found: C, 66.76; H, 5.92.

6.4.9. *N*-[5-Bromo-2-hydroxy-3-(adamantan-1-yl)-benzyl]acetamide (12). A finely pulverized mixture of 2-(1-adamantanyl)-4-bromophenol (**7**) (500 mg, 163 mmol) and N-(hydroxymethyl)acetamide (145 mg, 1.63 mmol) was added portionwise, at 10 °C while stirring, to 1.63 mL of a solution of CH_3COOH/H_2SO_4 concd 9:1. After the mixture was stirred at room temperature for 5 days, ice was added and the yellow solid was filtered and washed with water. Purification by flash chromatography (hexane/ethyl acetate 60:40) gave 225 mg (37%) of the pure product, mp 216 °C ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.77 (6H, s, 6Ad), 2.04 (3H, s, 3Ad), 2.05 (3H, s, CH_3), 2.10 (6H, s, 6Ad), 4.25 (2H, d, CH_2N, J = 6.8 Hz), 6.28 (1H, t, NH, J = 6.8 Hz), 7.05 (1H, d, 1Ar, J = 2.6 Hz), 7.23 (1H, d, 1Ar, J = 2.6 Hz). Anal. calcd for $C_{19}H_{24}BrNO_2$: C, 60.32; H. 6.39; N. 3.70. Found: C, 60.45; H, 6.32; N, 3.75.

6.4.10. *E*-3-[3′-(Acetylaminomethyl)-4′-hydroxy-5′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid (13). An amount of 195 mg (0.516 mmol) of **12** was reacted with methyl *p*-bromocinnamate according to the general procedure A. Purification by flash chromatography (ethyl acetate/dichloromethane 9:1) afforded 103 mg (43%) of 3-[3′-(acetylaminomethyl)-4′-hydroxy-5′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid methyl ester, mp 163 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.80 (6H, s, 6Ad), 2.05 (3H, s, OAc), 2.08 (3H, s, 3Ad), 2.20 (6H, s, 6Ad), 3.80 (3H, s, OMe), 4.40 (2H, d, CH_2N, J = 6.0 Hz), 6.32 (1H, t, NH, J = 6.0 Hz), 6.69 (1H, d, CH═, J = 15.8 Hz), 7.19 (1H, d, 1Ar, J = 2.6 Hz), 7.44 (1H, d, 1Ar, J = 2.6 Hz), 7.54–7.58 (4H, m, 4Ar), 7.72 (1H, d, CH═, J = 15.8 Hz).

The above ester (100 mg, 0.218 mmol) was hydrolyzed as described in the general procedure B, to obtain 70 mg (72%) of the pure product, mp 264 °C. ^{1}H NMR (300 MHz, acetone-d_6) δ: 1.83 (6H, s, 6Ad), 2.07 (3H, s, 3Ad), 2.25 (6H, s, 6Ad), 4.36 (2H, d, CH_2N, J = 6.0 Hz), 6.53 (1H, d, CH═, J = 15.8 Hz), 7.40 (1H, d, 1Ar, J = 2.3 Hz), 7.48 (1H, d, 1Ar, J = 2.3 Hz), 7.64 (2H, d, 2Ar, J = 8.3 Hz), 7.69 (1H, d, CH═, J = 15.8 Hz), 7.73 (2H, d, 2Ar, J = 8.3 Hz), 8.37 (1H, t, NH, J = 6.0 Hz), 10.41 (1H, s). MS (EI) *m*/*z* 445 (55, M+), 386 (100).

6.4.11. *N*-[5-Bromo-2-hydroxy-3-(adamantan-1-yl)-benzyl]-2-chloroacetamide (14). A finely pulverized mixture of 2-(adamantan-1-yl)-4-bromophenol (**7**) (500 mg,

163 mmol) and 2-chloro-*N*-(hydroxymethyl)acetamide (201 mg, 1.63 mmol) was added portionwise, at 10 °C while stirring, to 1.63 mL of a solution of CH_3COOH/H_2SO_4 concd 9:1. After the mixture was stirred at room temperature for 20 h, ice was added and the white solid was filtered and washed with water, to obtain 586 mg (87%) of the pure product, mp 154 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.75 (6H, s, 6Ad), 2.10 (9H, s, 9Ad), 4.10 (2H, s, CH_2Cl), 4.35 (2H, d, CH_2N, J = 6.0 Hz), 7.13 (1H, d, 1Ar, J = 2.3 Hz), 7.25 (1H, d, 1Ar, J = 2.3 Hz), 7.35 (1H, t, NH, J = 6.0 Hz), 9.0 (1H, br s, OH). Anal. calcd for $C_{19}H_{23}BrClNO_2$: C, 55.29; H, 5.62; N, 3.39. Found: C, 55.40; H, 5.49; N, 3.49.

6.4.12. *E*-3-{3′-[(2-Chloroacetylamino)-methyl]-4′-hydroxy-5′-(adamantan-1-yl)biphenyl-4-yl}-acrylic acid (15). An amount of 3.25 g (7.87 mmol) of **14** was reacted with methyl 4-bromocinnamate according to the general procedure C. Purification by flash chromatography (dichloromethane) gave 866 mg (27%) of 3-{3′-[(2-chloroacetylamino)-methyl]-4′-hydroxy-5′-(adamantan-1-yl)biphenyl-4-yl}-acrylic acid methyl ester, mp 127 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.78 (6H, s, 6Ad), 2.08 (3H, s, 3Ad), 2.19 (6H, s, 6Ad), 3.82 (3H, s, OMe), 4.13 (2H, s, CH_2Cl), 4.47 (2H, d, CH_2N, J = 6.0 Hz), 6.44 (1H, d, CH═, J = 15.0 Hz), 7.25 (1H, d, 1Ar, J = 2.3 Hz), 7.40 (1H, t, NH, J = 6.0 Hz), 7.45 (1H, d, 1Ar, J = 2.3 Hz), 7.54–7.60 (4H, m, 4Ar), 7.72 (1H, d, CH═, J = 15.0 Hz), 9.07 (1H, s, OH).

To a solution of 850 mg (1.72 mmol) of the above ester in 34 mL of dioxane, 10.2 mL of concentrated HCl was added. The solution was refluxed for 23 h. The solvent was evaporated, the residue taken up with water, and the solid formed was filtered and dried, to obtain 700 mg (92%) of the pure product, mp >300 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.72 (6H, s, 6Ad), 2.03 (3H, s, 3Ad), 2.14 (6H, s, 6Ad), 4.09 (2H, q, CH_2N, J = 5.3 Hz), 6.54 (1H, d, CH═ J = 15.8 Hz), 7.43 (1H, d, 1Ar, J = 2.3 Hz), 7.56 (1H, d, 1Ar, J = 2.3 Hz), 7.58 (1H, d, CH═, J = 15.8 Hz), 7.64 (2H, d, 2Ar, J = 7.1 Hz), 7.73 (2H, d, 2Ar, J = 7.1 Hz), 8.15 (3H, br s), 8.92 (1H, s). Anal. calcd for $C_{26}H_{30}ClNO_3$: C, 70.98; H, 6.87; N, 3.18. Found: C, 71.09; H, 6.80; N, 3.10.

6.4.13. 4′-Bromo-4-hydroxy-5-(adamantan-1-yl)biphenyl-3-carbaldehyde (17). To a solution of 2 g (5.22 mmol) of 4-(4′-bromophenyl)-2-(adamantan-1-yl)-phenol (**16**) and 1.12 g (1.22 mL, 10.44 mmol) of 2,6-lutidine in 11 mL of freshly distilled toluene, 0.68 g (2.61 mmol, 0.3 mL) of $SnCl_4$ was added over 10 min under a nitrogen atmosphere. A pale yellow precipitate was formed. The mixture was allowed to stir at room temperature for 20 min, then solid paraformaldehyde (0.626 g, 20.88 mmol) was added in one portion and the reaction mixture was stirred for additional 10 min, then heated at 90–95 °C for 7 h. After having cooled at room temperature, 30 mL of water and 6 mL of HCl 1 M were added. The aqueous phase was extracted with ethyl acetate, the extract washed with brine, dried over Na_2SO_4, and evaporated. The crude product was purified by flash chromatography (hexane/ethyl acetate 95:5) to give 1.3 g (60%) of the title compound, mp 189 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.8 (6H, s, Ad), 1.65 (3H, s, Ad), 2.19 (6H, s, Ad), 7.40 (2H, d, 2Ar, J = 8.3 Hz), 7.54 (2H, d, 2Ar, J = 8.3 Hz), 7.56 (1H, d, 1Ar, J = 2.3 Hz), 7.64 (1H, d, 1Ar, J = 2.3 Hz), 9.94 (1H, s, OH), 11.8 (1H, s, CHO). Anal. calcd for $C_{23}H_{23}BrO_2$: C, 67.16; H, 5.64. Found: C, 67.22; H, 5.73.

6.4.14. 4′-Bromo-3-hydroxymethyl-5-(adamantan-1-yl)-biphenyl-4-ol (18). To a solution of **16** (300 mg, 0.782 mmol) and 2,6-lutidine (0.799 mg, 0.872μl) in 1.72 mL of freshly distilled toluene 0.219 mL (1.87 mmol) were added with a syringe. The reaction mixture turned yellow with a yellow precipitate. The mixture was stirred at room temperature for 20 min, then solid paraformaldehyde (93.8 mg, 3.182 mmol) was added and the reaction mixture was stirred for additional 20 min, then heated at 95 °C for 5 h. After cooling at room temperature, water and 1 N HCl were added. The aqueous phase was extracted with ethyl acetate, the extract washed with brine, dried over Na_2SO_4, and evaporated. The crude product was purified by flash chromatography (hexane/ethyl acetate 85:15) to give 110 mg (34%) of the title compound, mp 178 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.83 (6H, s, 6Ad), 2.16 (3H, s, 3Ad), 2.24 (6H, s, 6Ad), 4.93 (2H, s, CH_2OH), 7.08 (1H, d, 1Ar, J = 1.6 Hz), 7.38 (1H, d, 1Ar, J = 1.6 Hz), 7.40 (2H, d, 2Ar, J = 8.2 Hz), 7.53 (2H, d, 2Ar, J = 8.2 Hz). Anal. calcd for $C_{23}H_{25}BrO_2$: C, 66.83; H, 6.10. Found: 66.96; H, 6.09.

6.4.15. 4′-Bromo-5-(adamantan-1-yl)biphenyl-3,4-diol (19). An amount of 300 mg (0.73 mmol) of compound **17** was dissolved in 3 mL of dry dichloromethane and then 180 mg (0.73 mmol) of 3-chloroperoxybenzoic acid was added. The mixture was refluxed for 20 h. The solvent was evaporated, the residue was dissolved in ethyl acetate, washed twice with $NaHCO_3$ (20%), brine, and dried over Na_2SO_4. Purification by flash chromatography (hexane/ethyl acetate 90:10) afforded 155 mg (53%) of the pure product, mp 178 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.78 (6H, s, 6Ad), 2.10 (3H,s, 3Ad), 2.15 (6H, s, 6Ad), 6.90 (1H, d, 1Ar, J = 2.3 Hz), 7.00 (1H, d, 1Ar, J = 2.3 Hz), 7.35 (2H, d, 2Ar, J = 8.3 Hz), 7.48(2H, d, 2Ar, J = 8.3 Hz). Anal. calcd for $C_{22}H_{23}BrO_2$: C, 66.17; H, 5.81. Found: C, 66.30; H, 5.97.

6.4.16. *E*-3-[3′,4′-Dimethoxy-5′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid (20). An amount of 150 mg (0.376 mmol) of compound **19** was reacted with methyl acrylate (51.7 mg, 0.602 mmol) according to the general procedure A. The crude product was purified by flash chromatography (dichloromethane/methanol 97:3) to give 127 mg (84%) of the highly unstable 3-[3′,4′-dihydroxy-5′-(1-adamantyl)-biphenyl-4-yl]-acrylic acid methyl ester, mp 208 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.79 (6H, s, 6Ad), 2.10 (3H,s, 3Ad), 2.20 (6H, s, 6Ad), 3.81 (3H, s, OMe), 6.42 (1H, d, CH═, J = 16.0 Hz), 6.98 (1H, d, 1Ar, J = 2.3 Hz), 7.05 (1H, d, 1Ar, J = 2.3 Hz), 7.48 (4H, m, 4Ar), 7.70(1H, d, CH═, J = 16.0 Hz).

To a suspension of sodium hydride (60% in mineral oil, 14 mg, 0.360 mmol) in 0.4 mL of DMF, 50 mg (0.124 mmol) of the above ester was slowly added, while maintaining the temperature at 20 °C. The mixture was then stirred at room temperature for 1 h, then methyl iodide (100 μL, 0.322 mmol) was added. After stirring for an additional hour, the mixture was poured into water and extracted with ethyl acetate. The organic phase was dried over Na_2SO_4, filtered, and evaporated. The residue was purified by flash chromatography (hexane: ethyl acetate 85:15) to give 10 mg (19%) of 3-[3′,4′-dimethoxy-5′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid methyl ester, mp 117 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.79 (6H, s, 6Ad), 2.10 (3H,s, 3Ad), 2.14 (6H, s, 6Ad), 3.81 (3H, s, OMe), 3.91 (6H, s, 2 OMe), 6.45 (1H, d, CH═, J = 15.7 Hz), 7.00 (1H, d, 1Ar, J = 2.2 Hz), 7.08 (1H, d, 1Ar, J = 2.2 Hz), 7.57 (4H, m, 4Ar), 7.70 (1H, d, CH═, J = 16.0 Hz), MS (EI) m/z 432 (M^+).

The above ester (24 mg, 0.055 mmol) was hydrolyzed according to the general procedure B for 4 days in the dark. The precipitated solid was filtered to give 20 mg (87%) of the pure product, mp 215 °C, ^{1}H NMR (300 MHz, acetone-d_6) δ: 1.83 (6H, s, 6Ad), 2.08 (3H,s, 3Ad), 2.16 (6H, s, 6Ad), 3.90 (3H, s, OMe), 3.96 (3H, s, OMe), 6.55 (1H, d, CH═, J = 15.8 Hz), 7.17 (1H, d, 1Ar, J = 2.2 Hz), 7.25 (1H, d, 1Ar, J = 2.2 Hz), 7.70–7.98 (4H, m, 4Ar), 7.93 (1H, d, CH═, J = 15.8 Hz). MS (EI) m/z 418 (M^+).

6.4.17. *E*-3-[4′-Hydroxy-3′-hydroxymethyl-5′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid (21). An amount of 100 mg (0.242 mmol) of **18** was reacted with methyl acrylate (33.3 mg, 0.387 mmol) as described previously (B). The residue was purified by flash chromatography (hexane/ethyl acetate 70: 30) to give 33 mg (33%) of the pure 3-[4′-hydroxy-3′-hydroxymethyl-5′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid methyl ester, mp 205 °C, ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.92 (6H, s, 6Ad), 2.13 (3H,s, 3Ad), 2.20 (6H, s, 6Ad), 3.95 (3H, s, OMe), 4.90(2H, s, CH_2OH), 6.43 (1H, d, CH═, J = 16.0 Hz), 7.10 (1H, d, 1Ar, J = 1.6 Hz), 7.42 (1H, d, 1Ar, J = 1.6 Hz), 7.53–7.60 (4H, m, 4Ar), 7.70 (1H, d, CH═, J = 16.0 Hz), 7.94 (1H, s).

The above ester (33 mg, 0.0789 mmol) was hydrolyzed as described before (B) to obtain 29 mg (91%) of the pure acid, mp > 300 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.73 (6H, s, 6Ad), 2.03 (3H,s, 3Ad), 2.14 (6H, s, 6Ad), 5.88 (1H, t, –OH, J = 5.3 Hz), 4.66 (2H, d, CH_2OH, J = 5.3 Hz), 6.50 (1H, d, CH═, J = 15.7 Hz), 7.30 (1H, d, 1Ar, J = 1.6 Hz), 7.35(1H, d, 1Ar, J = 1.6 Hz), 7.58 (1H, d, CH═, J = 15.7 Hz), 7.60 (2H, d, 2Ar, J = 8.3 Hz), 7.68 (2H, d, 2Ar, J = 8.3 Hz), 8.85 (1H, s, OH) 12.3 (1H, br s, COOH). MS (EI) m/z 404 (30), 386 (100), 305 (70), 240 (57), 165 (20).

6.4.18. *E*-3-[3′-Formyl-4′-hydroxy-5′-(adamantan-1-yl)-biphenyl-4-yl]-acrylic acid (22). Compound **17** was subjected to Heck condensation as described in general procedure A. The pure 3-(3′-formyl-4′-hydroxy-5′-(adamantan-1-yl)biphenyl-4-yl)-acrylic acid methyl ester was obtained after purification by flash chromatography (hexane/ethyl acetate 3:7) and crystallization from diisopropylether. Yield 66%, mp 245 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.56 (6H, s, Ad), 2.07 (3H, s, Ad), 2.20 (6H, s, Ad), 3.58 (3H, s, OMe), 6.46 (1H, d, CH═, J = 15.0 Hz), 7.59 (m, 4H, 4Ar), 7.60 (1H, d, 1Ar, J = 2.3 Hz), 7.70 (1H, d, 1Ar, J = 2.3 Hz), 7.73 (1H, d, CH═, J = 15.0 Hz), 9.95 (1H, s, OH), 11.9 (1H, s, CHO).

The above ester was hydrolyzed as described in general procedure B to obtain the corresponding acid, yield 83%, mp > 300 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.54 (6H, s, Ad), 2.08 (3H, s, Ad), 2.16 (6H, s, Ad), 6.55 (1H, d, CH═, J = 16.0 Hz), 7.59 (1H, d, CH═, J = 16.0 Hz), 7.66–8.18 (5H, m, 5Ar), 8.03 (1H, d, 1Ar, J = 2.3 Hz), 10.05 (1H, s, CHO), 12.05 (1H, s, OH). MS (EI) m/z 402 (M^+).

6.4.19. 4′-Bromo-4-methoxy-5-(adamantan-1-yl)biphenyl-3-ol (23). A mixture of **17** (345 mg, 0.839 mmol), MeI (502 mg, 3.54 mmol), and K_2CO_3 (167 mg, 1.21 mmol) in dry acetone (15 mL) was refluxed for 6 h. The solvent was evaporated, the residue was suspended in water and extracted with ethyl acetate. The organic phase was dried over Na_2SO_4, filtered, and evaporated to give 322 mg (90%) of 4′-bromo-4-methoxy-5-(adamantan-1-yl)biphenyl-3-carbaldehyde, mp. 145 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.75 (6H, s, Ad), 2.14 (9H, s, Ad), 3.72 (3H, s, OMe), 7.19 (2H, d, 2Ar, J = 8.3 Hz), 7.54 (2H, d, 2Ar, J = 8.3 Hz), 7.69 (1H, d, 1Ar, J = 1.9 Hz), 7.87 (1H, d, 1Ar, J = 1.9 Hz), 10.2 (1H, s, CHO).

To 220 mg (0.517 mmol) of the above aldehyde dissolved in 2 mL of dry dichloromethane, 354 mg (1.034 mmol) of 3-chloroperoxybenzoic acid (70%) was added portionwise. The solution was refluxed for 1 h. A yellow solid formed during the reaction. Dichloromethane was removed and the residue was dissolved in ethyl acetate, washed twice with $NaHCO_3$ (20%), with brine, and then dried over Na_2SO_4. Purification by flash chromatography (hexane/ethyl acetate 97:3) gave 66 mg (21%) of the phenol **23** and 85 mg (26%) of formic acid 5-adamantan-1-yl-4′-bromo-4-methoxy-biphenyl-3-yl ester, mp 103 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.55 (6H, s, Ad), 2.12 (9H, s, Ad), 3.87 (3H, s, OMe), 7.15 (1H, d, 1Ar, J = 2.3 Hz), 7.33 (1H, d, 1Ar, J = 2.3 Hz), 7.38 (2H, d, 2Ar, J = 8.3 Hz), 7.55 (2H, d, 2Ar, J = 8.3 Hz), 8.30 (1H, s).

A solution of 80 mg (0.181 mmol) of the above compound in 4 mL of methanol was treated with 100 μL of 10% KOH. After stirring for 1 h, the solvent was removed, the residue was suspended in water and acidified with 10% HCl. The formed yellow solid was filtered and dried. The title compound was obtained quantitatively (75 mg). ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.78 (6H, s, Ad), 2.12 (9H, s, Ad), 3.85 (3H, s, OMe), 7.00 (2H, d, 2Ar, J = 2.2 Hz), 7.40 (2H, d, 2Ar, J = 8.3 Hz), 7.55 (2H, d, 2Ar, J = 8.3 Hz), 8.30 (1H, s). Anal. calcd for $C_{23}H_{25}BrO_2$: C, 66.83; H, 6.10. Found: C, 67.00; H, 6.01.

6.4.20. *E*-3-[3′-Hydroxy-4′-methoxy-5′-(adamantan-1-yl)biphenyl-4-yl]-acrylic acid (24). Compound **23** (125 mg, 0.302 mmol) was reacted with methyl acrylate (41.5 mg, 0.483 mmol) according to the general procedure A. Purification by flash chromatography (hexane/ethyl acetate 80:20) afforded 84 mg (67%) of 3-(3′-hydroxy-4′-methoxy-5′-[(adamantan-1-yl)-biphenyl-4-yl]-acrylic acid methyl ester, mp 197 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.80 (6H, s, Ad), 2.11 (9H, s, Ad), 3.82 (3H, s, OMe), 6.45 (1H, d, CH═, J = 15.8 Hz), 7.08 (2H, d, 2Ar, J = 2.2 Hz), 7.55 (4H, m, 4 Ar), 7.70 (1H, d, CH═, J = 15.8 Hz).

The above ester (57 mg, 0.136 mmol) was treated with 7.8 mL of a 0.7 N NaOH solution in methanol under reflux for 2 h. After evaporation of methanol and addition of water, the mixture was acidified with 6 N HCl and the solid was filtered. Purification by flash chromatography (ethyl acetate/hexane 95:5) gave 10 mg (20%) of the pure product, mp >300 °C, ^{1}H NMR (300 MHz, acetone-d_6) δ: 1.83 (6H, s, Ad), 2.08 (3H, s, 3Ad), 2.28 (6H, s, 6Ad), 3.92 (3H, s, OMe), 6.55 (1H, d, CH═, J = 15.0 Hz), 7.08 (1H, d, 1Ar, J = 2.3 Hz), 7.14 (1H, d, 1Ar, J = 2.3 Hz), 7.67 (2H, d, 2Ar, J = 8.3 Hz), 7.70 (1H, d, CH═, J = 15.0 Hz), 7.72 (2H, d, 2Ar, J = 8.3 Hz). Anal. calcd for $C_{26}H_{28}O_4$: C, 77.20; H, 6.98. Found: C, 77.26; H, 7.07.

6.4.21. 4-Bromo-2-methoxy-6-(adamantan-1-yl)phenol (26). To a solution of 408 mg (1.97 mmol) of 4-bromoguaiacol (**25**) and 300 mg (1.97 mmol) of 1-adamantanol in 1 mL of dichloromethane, 108 mL (1.97 mmol) of concd sulfuric acid (98%) was added. The mixture was stirred for 1 h at room temperature, poured into water, neutralized with sodium bicarbonate, and extracted with dichloromethane.The organic phase was then dried over Na_2SO_4, filtered, and evaporated. The residue was purified by flash chromatography (hexane/ethyl acetate 95:5) to give 394 mg (60%) of the pure product, mp 159–160 °C, ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.78 (6H, s, 6Ad), 2.08 (9H, s, 9Ad), 3.85 (3H, s, OMe), 5.90 (1H, s), 6.85 (1H, d, 1Ar, J = 2.2 Hz), 6.93 (1H, d, 1Ar, J = 2.2 Hz). Anal. calcd for $C_{17}H_{21}BrO_2$: C, 60.54; H, 6.28. Found: C, 60.63; H, 6.19.

6.4.22. *E*-3-[4′-hydroxy-3′-methoxy-5′-(adamantan-1-yl]-biphenyl-4-yl]-acrylic acid (27). An amount of 200 mg (0.593 mmol) of compound **26** was reacted with methyl 4-bromocinnamate (287 mg, 1.13 mmol) as described in the general procedure C. Purification by flash chromatography (dichloromethane/hexane 50:50) gave 172 mg (70%) of 3-[4′-hydroxy-3′-methoxy-5′-(adamantan-1-yl)biphenyl-4-yl]acrylic acid methyl ester, mp 190 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.80 (6H, s, 6Ad), 2.08 (3H,s, 3Ad), 2.20 (6H, s, 6Ad), 3.81 (3H, s, OMe), 3.95 (3H, s, OMe), 6.08 (1H, s), 6.45 (1H, d, CH═, J = 16.0 Hz), 6.98 (1H, d, 1Ar, J = 1.9 Hz), 7.08 (1H, d, 1Ar, J = 1.9 Hz), 7.48–7.63 (4H, m, 4Ar), 7.73 (1H, d, CH═, J = 16.0 Hz), MS (EI) m/z 432 (M^+).

The above ester (90 mg, 0.215 mmol) was hydrolyzed as described above (B) to obtain 85 mg (98%) of the pure product, mp 218 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.73 (6H, s, 6Ad), 2.00 (3H,s, 3Ad), 2.12 (6H, s, 6Ad), 3.86 (3H, s, OMe), 6.50 (1H, d, CH═, J = 16.0 Hz), 7.00 (1H, d, 1Ar, J = 1.9 Hz), 7.58 (1H, d, CH═, J = 16.0 Hz), 7.63 (2H, d, J = 7.8 Hz), 7.70 (2H, d, J = 7.8 Hz), 8.54 (1H, s), 12.3 (1H, br s); MS (EI) m/z 404 (M^+).

6.4.23. 6-Bromo-4-(adamantan-1-yl)benzo[1,3]dioxole (29a). To a solution of 4-bromo-1,2-methylenedioxybenzene (**28a**) (1.5 g, 7.31 mmol) and 1-adamantanol (1.11 g, 7.31 mmol) in 3.7 mL of dichloromethane, 0.4 mL of concentrated sulfuric acid was added. The mixture was stirred for 4 h at room temperature, poured into water, neutralized with sodium bicarbonate, and extracted with dichloromethane. The solvent was dried and evaporated. Crystallization from diisopropylether gave 1.06 g (47%) of the desired compound, mp 133 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.80 (6H, s, 6Ad), 2.01 (6H, s, 6Ad), 2.12 (3H, s, 3Ad), 5.95 (2H, s, OCH2), 6.87 (2H, d, 2Ar, J = 2.8 Hz). Anal. calcd for $C_{17}H_{19}BrO_2$: C, 60.91; H, 5.71. Found: 61.02; H, 5.80.

6.4.24. 4-[7-(Adamantan-1-yl)benzo[1,3]dioxol-5-yl]benzaldehyde (30). To a solution of 875 mg (2.61 mmol) of **29a** in 5.3 mL of toluene, 2.61 mL (5.22 mmol) of a 2 N aqueous solution of Na_2CO_3, 90.5 mg (0.08 mmol) of $Pd(PPh_3)_4$, and 430 mg (2.87 mmol) of 4-formylbenzeneboronic acid (previously suspended in 1.2 mL of ethanol) were added. The mixture was refluxed under nitrogen for 7 h, then extracted with ethyl acetate. The organic phase was washed with brine, dried, and evaporated. The crude product was purified by flash chromatography (hexane/ethyl acetate 9:1) and crystallized from diethyl ether to give 658 mg (70%) of the desired aldehyde, mp 205 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.80 (6H, s, 6Ad), 2.09 (9H, s, 9Ad), 6.02 (2H, s, OCH_2), 7.01 (1H, d, 1Ar, J = 1.9 Hz), 7.04 (1H, d, 1Ar, J = 1.9 Hz), 7.68 (2H, d, 2Ar, J = 8.2 Hz), 7.92 (2H, d, 2Ar, J = 8.2 Hz), 10.07 (1H, s). Anal. calcd for $C_{24}H_{24}O_3$: C, 79.97; H, 6.71. Found: C, 80.12; H, 6.67.

6.4.25. *E*-3-{4-[7-(Adamantan-1-yl)benzo[1,3]dioxol-5-yl]-phenyl}-acrylic acid (31a). A solution of 300 mg (0.832 mmol) of **30** and 278 mg (0.832 mmol) of methoxycarbonylmethylenetriphenylphosphorane in 4.5 mL of chloroform was refluxed under nitrogen for 5 h. The solvent was evaporated and the residue was purified by flash chromatography (hexane/dichloromethane 45:55) to give 298 mg (86%) of 3-{4-[7-(adamantan-1-yl)benzo[1,3]dioxol-5-yl]-phenyl}-acrylic acid methyl ester, mp 205 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.72 (6 H, s, 6Ad), 2.06 (9H, s, 9Ad), 3.80 (3H, s, OMe), 5.97 (2H, s, OCH_2O), 6.44 (1H, d, CH═, J = 16.0 Hz), 6.95 (1H, d, 1Ar, J = 1.9 Hz), 6.98 (1H, d, 1Ar, J = 1.86 Hz), 7.52–7.60 (4H, m, 4Ar), 7.71 (1H, d, CH═, J = 16.0 Hz).

The above ester (200 mg, 0.48 mmol) was hydrolyzed as usual (B) to afford 150 mg (78%) of the pure product, mp > 300 °C, ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.72 (6H, s, 6Ad), 2.01 (9H, s, 9Ad), 6.01 (2H, s, OCH_2O), 6.52 (1H, d, CH═, J = 16.2 Hz), 6.99 (1H, d, 1Ar,

J = 1.9 Hz), 7.14 (1H, d, 1Ar, J = 1.9 Hz), 7.58 (1H, d, CH═, J = 16.2 Hz), 7.60 (2H, d, 2Ar, J = 8.5 Hz), 7.70 (2H, d, 2Ar, J = 8.5 Hz), 12.35 (1H, br s, COOH). Anal. calcd for $C_{26}H_{26}O_4$: C, 77.59; H, 6.51. Found: C, 77.64; H, 6.42.

6.4.26. 7-Bromo-5-(adamantan-1-yl)-2,3-dihydro-benzo-[1,4]dioxine (29b). To a solution of 4-bromo-1,2-ethylenedioxybenzene (500 mg, 2.28 mmol) (**28b**) and 1-adamantanol (347 mg, 2.28 mmol) in 1.15 mL of dichloromethane, 0.125 mL (2.28 mmol) of concentrated sulfuric acid was added. The mixture was stirred for 2 h at room temperature, poured into water, neutralized with sodium bicarbonate, and extracted with dichloromethane. The organic phase was evaporated to obtain a crude that was purified by flash chromatography (hexane/ethyl acetate 95:5) to give 280 mg (35%) of the pure product, mp 166–168 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.70 (6H, s, 6Ad), 2.00 (9H, s, 9Ad), 4.23 (4H, s, OCH_2), 6.74 (1H, d, 1Ar, J = 2.6 Hz), 6.88 (1H, d, 1Ar, J = 2.6 Hz).Anal. calcd for $C_{18}H_{21}BrO_2$: C, 61.90; H, 6.06. Found: C, 61.85; H, 6.01.

6.4.27. 3-{4-[8-(Adamantan-1-yl)-2,3-dihydro-benzo[1,4]-dioxin-6-yl]-phenyl}-acrylic acid (31b). Suzuki one-pot condensation between 200 mg (0.573 mmol) of compound **28b** with methyl 4-bromocinnamate was carried out as described in the general part (C). The residue was purified by flash chromatography (dichloromethane/hexane 1.1) to give 124 mg (50%) of 3-{4-[8-(adamantan-1-yl)-2,3-dihydro-benzo[1,4]dioxin-6-yl]-phenyl}-acrylic acid methyl ester, mp 209 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 1.78 (6H, s, 6Ad), 2.08 (3H, s, 3Ad), 2.13 (6H, s, 6Ad), 3.83 (3H, s, OMe), 4.33 (4H, s, OCH_2), 6.45 (1H, d, CH═, J = 16.0 Hz), 7.00 (1H, d, 1Ar, J = 2.3 Hz), 7.05 (1H, d, 1Ar, J = 2.3 Hz), 7.50–7.63 (4H, m, 4Ar), 7.70 (1H, d, CH═, J = 16.0 Hz).

The above ester (70 mg, 0.163 mmol) was hydrolyzed as usual (B) to afford 62 mg (91%) of the pure product, mp 288 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.71 (6H, s, 6Ad), 2.02 (3H, s, 3Ad), 2.08 (6H, s, 6Ad), 4.25 (4H, s, OCH_2), 6.50 (1H, d, CH═, J = 15.6 Hz), 6.97 (1H, d, 1Ar, J = 1.9 Hz), 7.02 (1H, d, 1Ar, J = 1.9 Hz), 7.56 (1H, d, CH═, J = 15.6 Hz), 7.58 (2H, d, 2Ar, J = 8.25 Hz), 7.69 (2H, d, 2Ar, J = 8.3 Hz). MS (EI) m/z 416 (M^+).

6.4.28. *Z*-2-Bromo-3-[4-(7-adamantan-1-yl)benzo[1,3]dioxol-5-yl)-phenyl]-acrylic acid ethyl ester (32) and *E*-2-bromo-3-[4-(7-adamantan-1-yl)benzo[1,3]dioxol-5-yl)-phenyl]-acrylic acid ethyl ester (33). A solution of 360 mg (0.999 mmol) of 30 and 472 mg (0.999 mmol) of ethoxycarbonylbromomethylenetriphenylphosphorane in 5.3 mL of chloroform was refluxed under nitrogen for 8 h. The solvent was evaporated and the crude product was purified by flash chromatography (hexane/dichloromethane 45:55) to give 369 mg (72%) of the Z isomer (32), mp 150 °C, and 82 mg (16%) of the E isomer (33), mp 188 °C. ^{1}H NMR Z isomer (300 MHz, $CDCl_3$) δ: 1.39 (3H, t, Et, J = 7.5 Hz), 1.80 (6H, s, 6Ad), 2.09 (9H, s, 9Ad), 4.37 (2H, q, Et, J = 7.5 Hz), 5.98 (2H, s, CH_2O), 6.97 (1H, d, 1Ar, J = 1.9 Hz), 7.00 (1H, d, 1Ar, J = 1.9 Hz), 7.56 (2H, d, 2Ar, J = 8.3 Hz), 7.90 (2H, d, 2Ar, J = 8.3 Hz), 8.22 (1H, s, CH═). Anal. calcd for $C_{28}H_{29}BrO_4$: C, 66.01; H, 5.74. Found: C, 66.18; H, 5.54.

^{1}H NMR E isomer (300 MHz, $CDCl_3$) δ: 1.20 (3H, t, Et, J = 7.5 Hz), 1.75 (6H, s, 6Ad), 2.02 (9H, s, 9Ad), 4.24 (2H, q, Et, J = 7.5 Hz), 5.97 (2H, s, CH_2O), 6.92 (1H, d, 1Ar, J = 1.8 Hz), 6.98 (1H, d, 1Ar, J = 1.8 Hz), 7.31 (2H, d, 2Ar, J = 8.3 Hz), 7.38 (1H, s, CH═), 7.49 (2H, d, 2Ar, J = 8.3 Hz). Anal. calcd for $C_{28}H_{29}BrO_4$: C, 66.01; H, 5.74. Found: C, 66.12; H, 5.66.

6.4.29. {4-[7-(Adamantan-1-yl)benzo[1,3]dioxol-5-yl]-phenyl}-propynoic acid (34). A suspension of 350 mg (0.687 mmol) of the ester **32** in 2.4 mL of a 25% solution of potassium hydroxide in methanol was refluxed for 1 h. The solvent was evaporated and water was added to the residue, then HCl 12 N was slowly added. The precipitated solid was filtered and dried. Crystallization from diisopropylether gave 109 mg (40%) of the desired compound, mp 204 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.76 (6H, s, 6Ad), 2.03(9H, s, 9Ad), 6.02 (2H, s, CH_2O), 6.98 (1H, d, 1Ar, J = 1.9 Hz), 7.14 (1H, d, 1Ar, J = 1.9 Hz), 7.54 (2H, d, 2Ar, J = 8.3 Hz), 7.64 (2H, d, 2Ar, J = 8.3 Hz). MS (EI) m/z 356 ($M-CO_2$).

6.4.30. *E*-2-Bromo-3-[4-(7-adamantan-1-yl)benzo[1,3]dioxol-5-yl)-phenyl]-acrylic acid (35). The ester **33** (73 mg, 0.143 mmol) was hydrolyzed as usual (B) to afford 57 mg (89%) of the pure product, mp 239 °C, ^{1}H NMR (300 MHz, DMSO-d_6) δ: 1.72 (6H, s, 6Ad), 2.00 (9H, s, 9Ad), 5.98 (2H, s, OCH_2), 6.95 (1H, d, 1Ar, J = 1.2 Hz), 7.15 (1H, d, 1Ar, J = 1.2 Hz), 7.31 (1H, s, CH═), 7.39 (2H, d, 2Ar, J = 8.5 Hz), 7.56 (2H, d, 2Ar, J = 8.5 Hz), 13.4 (1H, br s). Anal. calcd for $C_{26}H_{25}BrO_4$: C, 64.87; H, 5.23. Found: C, 64.98; H, 5.12.

6.5. Cellular sensitivity to drugs

In ovarian carcinoma cells, cellular sensitivity to drugs was evaluated by growth-inhibition assay after 72-h drug exposure. Cells in the logarithmic phase of growth were seeded in duplicate into 6-well plates. Twenty-four hours after seeding, the drug was added to the medium. Cells were harvested 72 h after drug exposure and counted with a cell counter. In leukemia cells, drug exposure was 24 h and the growth inhibition was assessed after 72 h. IC_{50} is defined as the drug concentration causing a 50% reduction of cell number compared with that of untreated control.

6.6. Determination of apoptosis

Apoptosis was determined in ovarian carcinoma IGROV-1 cells by TUNEL assay following 72 h-exposure to the drug.[8] Treated cells were fixed in 4% paraformaldehyde, for 60 min, at room temperature, washed and resuspended in ice-cold PBS. The in situ cell death detection kit fluorescein (Roche, Mannheim, Germany) was used according to the manufacturers' instructions

and the samples were analyzed by flow cytometry (Becton Dickinson).

6.7. Cell cycle analysis

The cell cycle distribution was analyzed in propidium iodide-stained cells by FACScan flow cytometry, as described.[8]

6.8. Western blot analysis

Cells were treated for the indicated times with seven selected compounds (Fig. 2) at a cytotoxic concentration corresponding to IC_{80}. Analysis of each MAP protein and their activated (i.e., phosphorylated) form was performed as previously reported.[8]

6.9. Cytodifferentiation

The nitro blue tetrazolium (NBT) reduction assay[16] was performed on extracts of phorbol myristate acetate-stimulated cells and the percentage of NBT^+ cells was determined with a spectrophotometric assay at 540 nm. NB4 tumor cells were exposed at sub-toxic concentrations of the novel retinoids for 3 days.

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