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# Synthesis of novel steroid analogues containing nitrile and disulfide moieties via palladium-catalyzed cross-coupling reactions

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ABSTRACT

Grundmann's ketone (**3**) was used as a common building block for introduction of the C+D ring motif of steroids to prepare series of novel secosteroids containing nitrile or disulfide functionalities. Key steps in synthesis of these novel steroid analogues are palladium-catalyzed cross-coupling reactions. All target compounds were characterized by comprehensive analysis; in particular the stereochemical assignments of dinitriles **12a** and **12b** could be carried out in detail by NMR experiments. The pharmacological potential of all compounds was initially verified by a cytotoxicity test.



## 1. Introduction

Steroids constitute a highly important class of biologically active compounds that are widely used for therapeutic purposes. Further, steroids play pivotal roles in human metabolism by acting as hormones (androgens, estrogens, gestagens, glucocorticoids, mineralocorticoids) or essential parts of cell membranes (cholesterol). Introduction of new structural motifs into steroids frequently results in enhanced or completely different biological activities. In glucocorticoids an additional double bond (Δ1,2) results in enhanced potency and receptor selectivity,[1] whereas the potent antiandrogen and gestagen cyproterone acetate contains an additional cyclopropane ring annulated at the 1,2-bond.[1] Additional amino groups at ring A are found in the plakinamines, a class of marine steroid alkaloids with cytotoxic and antifungal activities,[2] and the 2-hydroxy-3-aminosteroid amafolone shows antiarrhythmic activity.[3] Among the large group of biologically active heterosteroids the 5α-reductase inhibitors finasteride and dutasteride (4-azasteroids) gained particular importance in the treatment of benign prostatic hyperplasia and alopecia.[4]

Steroids containing functional groups in ring A (and occasionally ring B) that are capable for reacting with nucleophilic functional groups, and thus binding covalently to biomolecules (enzymes) have gained considerable interest as cytotoxic and antimicrobial agents in the recent past. The withanolides,[5] a large class of oxidized sterols isolated from *Solanaceae* and other plants, show cytotoxic,[6,7] antifungal,[8] leishmanicidal, and trypanocidal[9] activities. The vast majority of bioactive withanolides contains an enone structure in ring A of the steroidal ring system, frequently accompanied by an epoxide group at ring B. Numerous bioactive oxidized sterols have been isolated from marine organisms,[10] and related semisynthetic epoxysterols exhibit potent cytotoxicity.[11] The metabolites of estradiol, both 2-hydroxy- and 4-hydroxyestradiol, can be further oxidized in vivo to give the corresponding *ortho*-quinones, which then undergo Michael-type additions with physiological nucleophiles like thiols and amino groups[12,13] and DNA bases like adenine.[14]

Recent investigations of our group revealed that steroid analogues containing a cyclohex-2-ene-1,4-dione Michael acceptor system in ring A exhibit outstanding cytotoxic activity.[15] In the course of these investigations we also prepared secosteroids containing Michael acceptor groups (*N*-substituted maleimides **1** and **2**; Fig. 1), which showed moderate cytotoxic activities.[15] This prompted us to investigate further secosteroid analogues containing functional groups that are capable of binding covalently to proteins, once again located at positions close to ring A of the native steroid backbone.

The most promising functional group for binding drugs to biomolecules is the thiol side chain of cysteine residues in proteins.[16,17] The thiol group can undergo nucleophilic addition to appropriate Michael acceptor systems (e.g., maleimides, enones) ending up in alkylthio-linked products (Fig. 2a).[18] Thiols are also able to bind covalently to nitrile groups under formation of thioimidates (Fig. 2b). In contrast to Michael additions, the thioimidate

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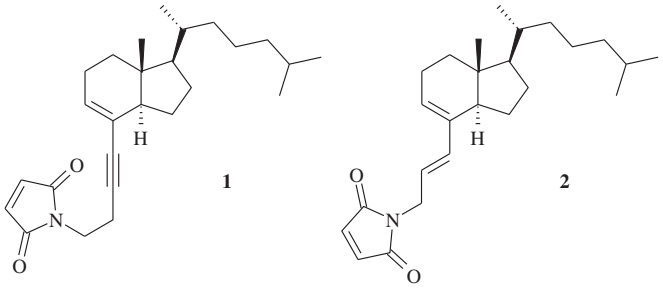


**Fig. 1.** Bioactive secosteroids containing maleimide functionalities.[15]

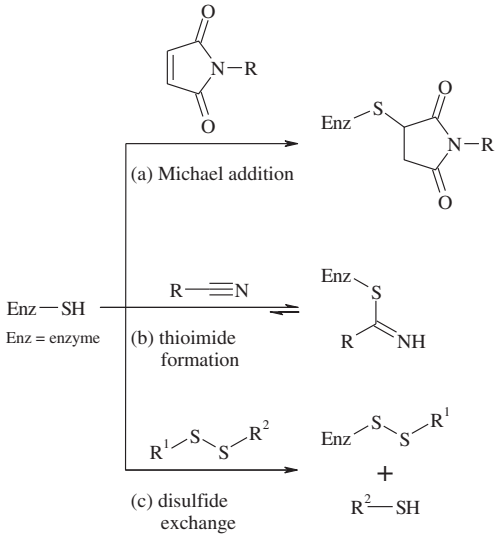


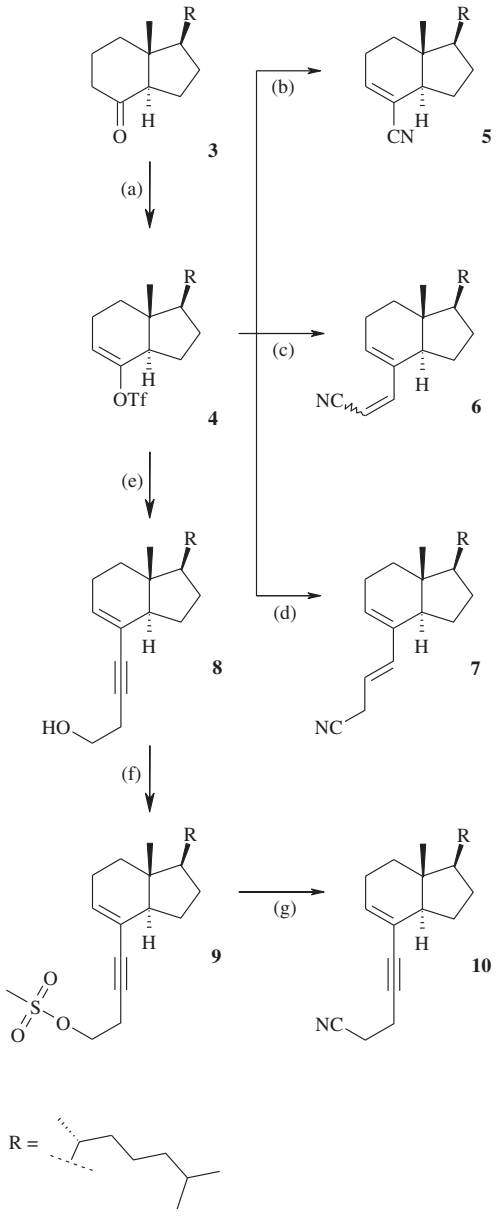

**Fig. 2.** Possible covalent enzyme–substrate binding modes.

formation is reversible in vivo.[19] Furthermore the thiol side chain of cysteines in proteins can undergo disulfide exchange reactions with symmetrical or unsymmetrical disulfides to bind thiol-derived active agents covalently (Fig. 2c).[20] In this communication we report on our investigations on analogues of the maleimide lead compounds **1** and **2** containing nitrile and disulfide moieties as reactive functional groups.

## 2. Results and discussion

### 2.1. Chemistry

Based on fundamental investigations of Alberola et al.[21] a series of analogues derived from Grundmann's ketone were prepared. These products are characterized by bearing a nitrile group attached to the bicyclic ring system directly a C-4 or separated by a $C_2$-, $C_3$- or $C_4$-spacer. For this purpose Grundmann's ketone (**3**), readily available by ozonolysis of vitamine $D_3$, was converted to the vinyl triflate **4** under kinetic control following a protocol worked out by De Riccardis et al.[22,23] Catalyzed by tetrakis(triphenylphosphine)-palladium(0) the vinyl triflate **4** was converted into the nitrile **5** using potassium cyanide and 18-crown-6. Heck reaction[24,25] of **4** and acrylonitrile catalyzed by tri-*o*-tolylphosphine and palladium(II) acetate under microwave irradiation gave an (*E*/*Z*) mixture of the cyanodiene **6** in moderate yield, which was not separable by column chromatography. In an analogous manner the Heck reaction was performed with but-3-enenitrile to obtain the nitrile **7** in moderate yield as shown in Scheme 1.

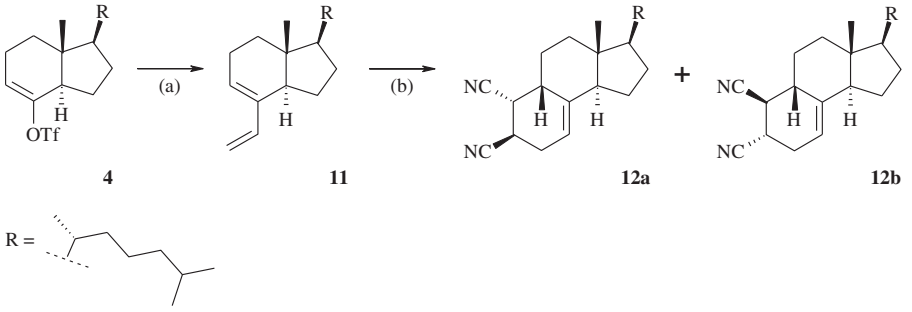



**Scheme 1.** Reagents and conditions: (a) sodium bis(trimethylsilyl)amide, THF, −78 °C, 1 h, then *N*-phenyl-bis(trifluoromethanesulfonimide), −78 °C to rt, 2.2 h (67%);[15] (b) potassium cyanide, tetrakis(triphenylphosphine)-palladium(0), 18-crown-6, toluene, 75 °C, 2 h (40%); (c) palladium(II) acetate, tri-*o*-tolylphosphine, acrylonitrile, acetonitrile, tributylamine, MW, 150 W, 150 °C, 10 min (54%); (d) palladium(II) acetate, tri-*o*-tolylphosphine, but-3-enenitrile, acetonitrile, tributylamine, MW, 150 W, 150 °C, 10 min (54%); (e) but-3-yn-1-ol, diethylamine, DMF, copper(I) iodide, bis(triphenylphosphine)palladium(II) dichloride, MW: 300 W, 120 °C, 5 min (98%);[15] (f) mesyl chloride, triethylamine, dichlormethane, 3 h, −10 °C (27%); (g) sodium cyanide, DMF, rt, 2 h (21%).

In contrast acetylenic alcohol **8** was obtained in excellent yield by Sonogashira coupling[26] of **4** and but-3-yn-1-ol catalyzed by bis(triphenylphosphine)-palladium(II) dichloride under microwave conditions. Reaction of alcohol **8** with mesyl chloride gave the mesylate **9** in poor yield. Finally, **9** was converted to the pent-4-ynenitrile **10** with sodium cyanide as shown in Scheme 1.

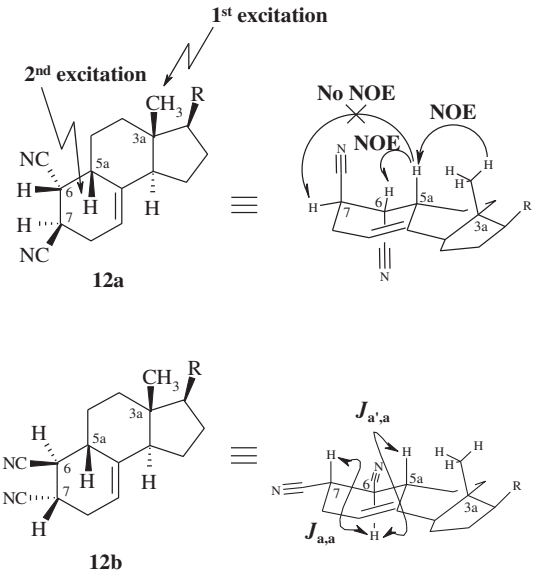
Stille coupling of the vinyl triflate **4** with vinyltributyltin, catalyzed by tetrakis(triphenylphosphine)-palladium(0), gave the 1,3-diene **11** in excellent yield. This diene had previously been converted to a 1,4-dioxocholestane by Diels–Alder cycloaddition with *p*-benzoquinone,[23] and to several moderately cytotoxic steroid-anthraquinone hybrids by cycloaddition/oxidation with

naphthoquinones by De Riccardis et al.[23] Using fumaronitrile ((*E*)-butenedinitrile) as dienophilic reactant, we obtained a separable 2:1 mixture of the isomeric dinitriles **12a** and **12b** (Scheme 2).




**Scheme 2.** Reagents and conditions: (a) tetrakis(triphenylphosphine)-palladium(0), lithium chloride, vinyltributyltin, THF, 75 °C, 2 h (98%);[15] (b) fumaronitrile, toluene, 120 °C, 12 h (**12a**: 42%, **12b**: 23%).

Due to the well-known absolute configuration at C-3a of starting material **3** stereochemical assignments of the dinitriles **12a** and **12b** derived thereof could be carried out by 1D DPFGSE-NOE experiments as well as by determination of *J*-coupling constants with the help of phase-sensitive double-quantum filtered COSY (DQF-COSY). The 1D NOE assignment of the absolute configuration of the newly formed stereogenic centers at C-5a, C-6, and C-7 of compound **12a** is presented in Scheme 3 and the resulting spectra are shown in Fig. 3. Selective excitation of the proton resonances of the angular methyl group attached to C-3a resulted in a NOE of the proton in position 5a. This clearly shows that the methyl group and 5a-H are located at the same side of the plane of the central ring. Hence, C-5a of compound **12a** is *S*-configured. A further DPFGSE-NOE experiment was performed to assign the absolute configuration of C-6. In the 1D NOE spectrum an NOE for 6-H was observed after selectively exciting 5a-H with a π-pulse. Hence, it can be deduced that 5a-H and 6-H are cis-orientated and C-6 is therefore *R*-configured. Since from mechanistic aspects the [4+2]-cycloaddition of the (*E*)-dinitrile fumaronitrile must lead to a *trans*-dinitrile,[27] the absolute stereochemistry of compound **12a** is proven unambiguously as shown in Scheme 3.




**Scheme 3.** Observed NOEs in the 1D DPFGSE-NOE experiments with **12a** in the upper panel and pseudo-axial–axial ($J_{a',a}$) and axial–axial ($J_{a,a}$) vicinal *J*-couplings of **12b** in the lower panel.

Due to partially overlapping resonances of 5a-H and 6-H of the isomeric dinitrile **12b** the stereochemical assignment could not be performed by NOE experiment in the same manner as for **12a** because the second excitation pulse on 5a-H cannot be considered as selective and therefore would lead to ambiguous results. Nevertheless, the first pulse on the 3a-$CH_3$ group could be performed selectively, resulting in an NOE of 5a-H revealing the 1,4-diaxial orientation of 3a-$CH_3$ and 5a-H. For this reason, we have decided to determine the absolute configuration by measurement of the coupling constants of 7-H to 6-H and of 6-H to 5a-H of **12b**. Simple quantum calculation with MM2 (Chem3D Ultra, Cambridge Soft®) revealed that the newly formed rings of **12a** as well as of **12b** adopt pseudo-chair conformations. If one assumes that C-6 and C-7 of **12b** have opposite configurations (6-*S* and 7-*S*, respectively) as compared to **12a**, then both protons at these stereogenic centers must have axial orientation. Since vicinal axial–axial coupling constants are typically larger than equatorial–axial and equatorial–equatorial coupling constants, it must be expected that in the resonance signals of 6-H and 7-H two axial–axial coupling constants each are embedded. To distinguish the also expected huge coupling of 7-H to the pseudo-axial proton at C-8 a phase-sensitive DQF-COSY experiment with high resolution was performed. The spectra yielding from this experiment show cross peaks with partially in-phase and anti-phase splitting. The anti-phase splitting originates from the active coupling, that is to say the coupling that creates the respective cross peak. The in-phase splitting gives rise to couplings to all other protons. Fig. 4 shows an excerpt of the obtained DQF-COSY spectrum and the extracted relevant 7-H/6-H cross peak is indicated by a rectangle. The cross peaks consists of an anti-phase and an in-phase splitting with a coupling of nearly the same magnitude (11.0 Hz) for the active coupling (7-H/6-H, green-red contours) and the passive coupling (6-H/5a-H red-red contours). The almost identical coupling constants generate the triplet of 6-H. The observed vicinal axial-axial and pseudo-axial-axial coupling constants confirm our stereochemical suggestion for compound **12b**. Noteworthy to say, that no such large vicinal couplings were observed in the signals for 6-H and 7-H in the $^1$H NMR spectrum of compound **12a**, due to the fact that both protons have equatorial orientation.

A second series of compounds, consisted of unsymmetrical disulfides, was synthesized starting from acetylenic alcohol **8**[15] as shown in Scheme 4. Mitsunobu reaction of **8** with thioacetic acid, diisopropyl azodicarboxylate, and triphenylphosphine gave the thioester **13** in excellent yield. Lithium aluminum hydride reduction of **13** gave the thiol **14**, which was subjected to disulfide exchange reactions with various symmetrical disulfides **15a**–**f**.

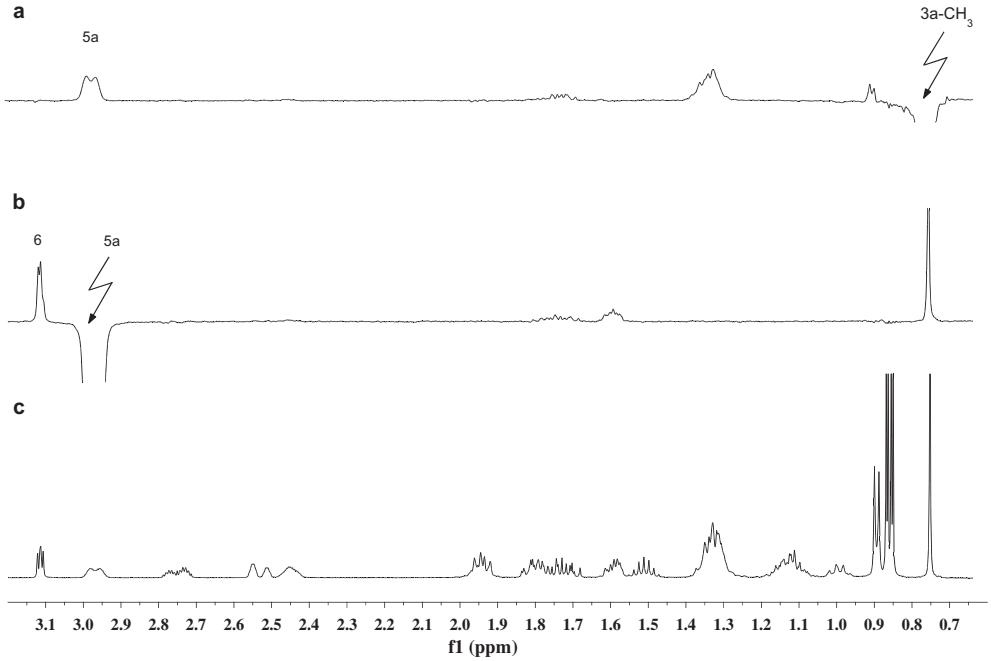



**Fig. 3.** (a) 1D DPFGSE-NOE with selective pulse on 3a-$CH_3$, (b) 1D DPFGSE-NOE with selective pulse on 5a-H, and (c) reference $^1$H NMR spectrum of compound **12a**.

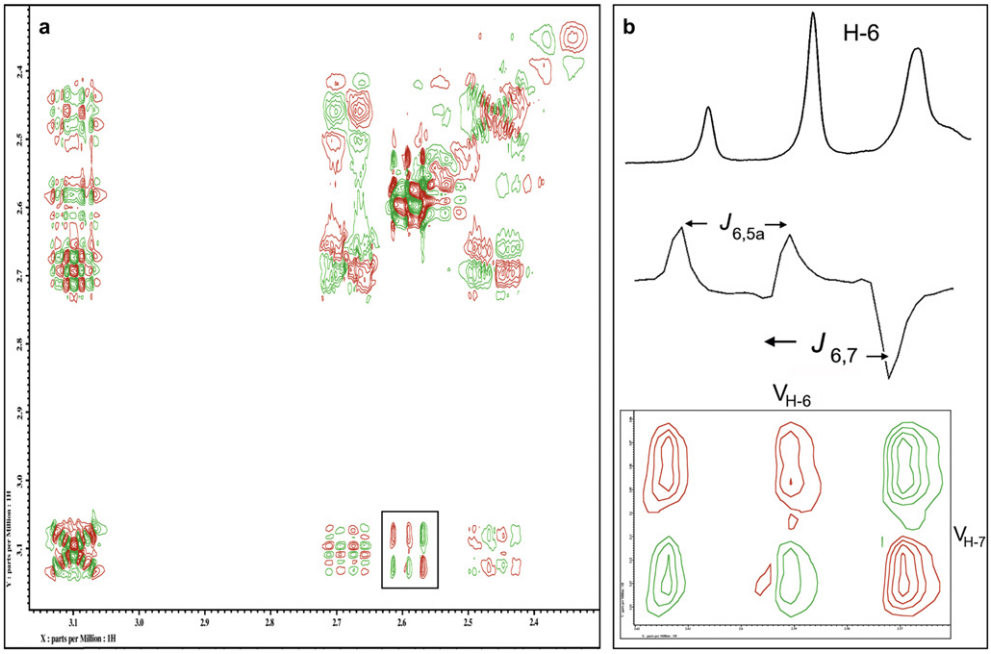



**Fig. 4.** (a) Excerpt of a 500 MHz phase-sensitive DQF-COSY spectrum of **12b** in $CDCl_3$ at 25 °C. Two transients, 4096×512 data matrix. (b) Extracted 7-H/6-H two-dimensional cross peak (lower panel). One-dimensional slice through *f2* of the respective cross peak (middle panel). Resonance signal of 6-H in the conventional $^1$H NMR spectrum of **12b** (upper panel). The outer peak in the up field is broadened due to overlapping with 5a-H.

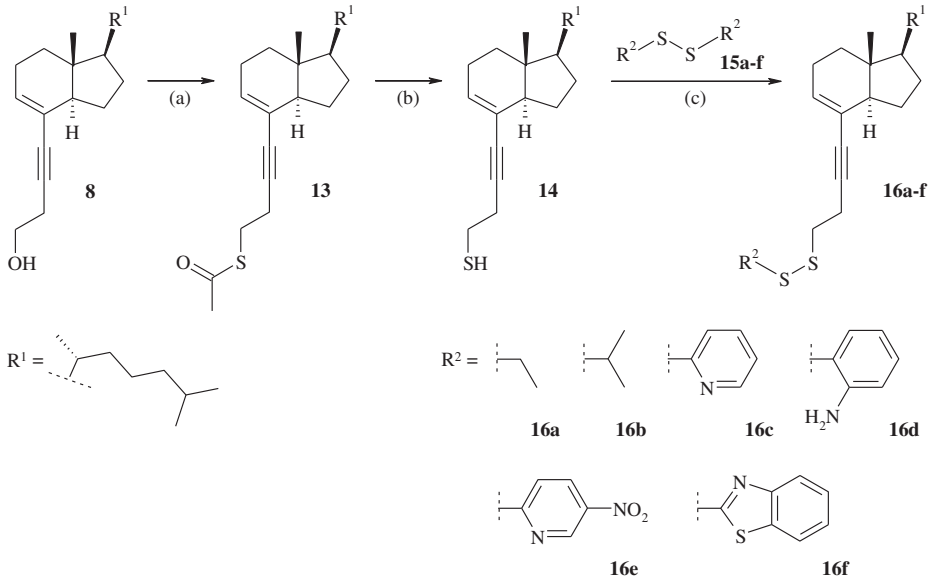
These reactions were quite sluggish and gave unsymmetrical disulfides **16a**–**f** containing aliphatic and heteroaromatic residues in poor to moderate yields.

### 2.2. Cytotoxic activity

A first screening for cytotoxic activities of the compounds was performed in an MTT test[28] on human leukemia HL-60 cells. Cisplatin was used as reference. The results are shown in Table 1. The mononitriles **5**, **6**, **7**, and **10** showed only very poor activities, even weaker than the acetylenic alcohol **8**. Surprisingly, the isomeric tricyclic dinitriles **12a** and **12b** differed significantly in their cytotoxic activities. While **12b** was completely inactive, its isomer **12a** showed an interesting $IC_{50}$ value of 12 μM. Among the disulfides **16a**–**f**, those containing aromatic residues (**16c**–**f**) showed moderate to good activities, whereas the aliphatic disulfides **16a** and **16b** are almost inactive.

## 3. Conclusion

A number of secosteroid derivatives of biological interest were prepared using palladium-catalyzed cross-coupling reactions as the key steps. Secosteroids **1** and **2**, which are Michael acceptors due to their *N*-alkylmaleimide moieties were used as lead structure for the




**Scheme 4.** Reagents and conditions: (a) diisopropyl azodicarboxylate, triphenylphosphine, thioacetic acid, THF, 0 °C to rt, 2 h (96%); (b) lithium aluminum hydride, diethyl ether, rt, 30 min, (93%); (c) symmetrical disulfide (**a**=diethyl disulfide, **b**=diisopropyl disulfide, **c**=2,2′-dithio dipyridine, **d**=bis(2-aminophenyl) disulfide, **e**=2,2′-dithiobis(5-nitropyridine), dichloromethane, rt, 12 h, (6–20%).

**Table 1**
Cytotoxic activities of investigated compounds against HL-60 cells determined in an MTT test

| Compound | Molecular formula | $IC_{50}$ [μM] |
|---|---|---|
| Cisplatin | $Cl_2H_6N_2Pt$ | 5 |
| **1**[15] | $C_{26}H_{37}NO_2$ | 46 |
| **2**[15] | $C_{25}H_{37}NO_2$ | 32 |
| **5** | $C_{19}H_{31}N$ | 27 |
| **6** | $C_{21}H_{33}N$ | 34 |
| **7** | $C_{22}H_{35}N$ | 34 |
| **8**[15] | $C_{22}H_{36}O$ | 13 |
| **9** | $C_{23}H_{38}O_{23}S$ | 54 |
| **10** | $C_{23}H_{35}N$ | 29 |
| **11**[15] | $C_{20}H_{34}$ | >100 |
| **12a** | $C_{24}H_{36}N_2$ | 12 |
| **12b** | $C_{24}H_{36}N_2$ | >100 |
| **16a** | $C_{24}H_{40}S_2$ | 40 |
| **16b** | $C_{25}H_{42}S_2$ | >100 |
| **16c** | $C_{27}H_{39}NS_2$ | 15 |
| **16d** | $C_{28}H_{41}NS_2$ | 16 |
| **16e** | $C_{27}H_{38}N_2O_2S_2$ | 11 |
| **16f** | $C_{29}H_{39}NS_3$ | 29 |

development of novel secosteroid analogues containing nitrile or disulfide functionalities. Like the lead compounds, both nitrile and disulfide analogues should be able to bind covalently to thiol residues of cellular target proteins. The vinyl triflate **4** derived from Grundmann's ketone (**3**), which itself is readily available from commercial vitamin $D_3$, was used as a common building block for introduction of the C+D ring motif of steroids. Palladium-catalyzed cyanation of **4** gave the nitrile **5**, whereas Heck olefinations with acrylonitrile and but-3-enenitrile led to cyanodiene **6** and cyanomethyldiene **7**. The acetylenic nitrile **10** was obtained via the Sonogashira product **8** and the corresponding mesylate **9**. Diels–Alder cycloaddition of the 1,3-diene **11**, accessible from vinyl triflate **4** via Stille coupling, with fumaronitrile gave two isomeric tricyclic dinitriles **12a** and **12b**, whose absolute configurations were determined by NMR experiments.

A second series of secosteroid analogues, containing unsymmetrical terminal disulfide moieties were prepared starting from the Sonogashira product **8**. The terminal hydroxy group was converted to a thiol in two steps, and subsequent disulfide exchange reactions with a number of symmetrical aliphatic and aromatic disulfides gave the desired unsymmetrical disulfides **16a-f** in moderate yields.

The screenings for cytotoxic activity revealed that a considerable number of the new steroid analogues show higher activity than the lead compounds **1** and **2**. In particular the cytotoxic activities of the isomeric tricyclic dinitriles **12a** and **12b** are remarkable, due to the clear difference of the screening results.

Further investigations are under way in order to elucidate the molecular mode of action of the most active compounds from this project.

## 4. Experimental

### 4.1. Chemistry

IR spectra were obtained on a Perkin Elmer FT-IR Paragon 1000 spectrometer. NMR spectra were recorded on JEOL JNM-Eclipse+400 (400 MHz) and JNM-Eclipse+500 (500 MHz) spectrometers with tetramethylsilane as an internal standard. All spectra were recorded in $CDCl_3$ as solvent and chemical shifts are reported in parts per million (ppm, $\delta$). *J* values are given in hertz. Signal assignments were carried out based on $^1H$, $^{13}C$, HSQC, HMQC, COSY, and HMBC spectra. DPFGSE-NOE pulse sequence was used from the JEOL pulse library. A Gaussian shaped pulse with 50 ms duration was applied to perform selective excitation. Mixing time was 1.0 s and a relaxation delay of 1.3 s was applied. Spectra were recorded with 32K points and 32 scans. Processing was performed with MestReNova v5.1.0 software and an exponential window function of 1.0 Hz was applied. Phase-sensitive DQF-COSY pulse sequence from the JEOL pulse library was used. The spectrum was recorded with 4096×512 data points with two scans per increment and eight dummy scans. Resulting data were analyzed using JEOL Delta v5.0 software. A sine-bell window function was applied and zero filling was performed to obtain a symmetrical data matrix. A slice along *f*2 was created using the automatic data slicer of the JEOL processing software. Mass spectra (MS) were run by chemical impact or electron impact (CI or EI) on a Hewlett Packard 5989 A mass mpectrometer with 59980B Particle Beam LC/MS interface. HRMS were performed by electron impact (EI) at 70 eV on a Jeol JMS

GCmate II. Anhydrous reactions were carried out in a nitrogen atmosphere. Solvents were of HPLC grade or p.a. grade, if not they were distilled before use. All chemicals were purchased from Sigma–Aldrich (Schnelldorf, Germany) and Acros Organics (Geel, Belgium). Microwave-promoted syntheses were performed on a single-mode microwave reactor (Discover) equipped with an IR temperature sensor from CEM (Kamp-Lintfort, Germany). Reactions were monitored by thin-layer chromatography (TLC) using precoated plastic sheets POLYGRAM® SIL G/UV254 from Macherey-Nagel (Düren, Germany). Compounds on TLC plates were detected under UV light at 254 nm and visualized by immersion in a solution of 5% $(NH_4)_6Mo_7O_{24}\cdot 4H_2O$, 0.2% $Ce(SO_4)_2\cdot 4H_2O$, and 5% concd $H_2SO_4$. Merck silica gel 60 was used as stationary phase for flash column chromatography. Purity of the synthesized compounds was >95% (HPLC).

*4.1.1. (1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-indene-4-carbonitrile (**5**).* Potassium cyanide (0.13 g; 2.00 mmol), tetrakis(trip-henylphosphine)-palladium(0) (35.0 mg; 30.0 μmol), and 18-crown-6 (20 mg; 0.076 mmol) were added to a solution of **4**[23] (0.39 g; 1.00 mmol) and toluene (3 mL). The reaction mixture was stirred 20 min at rt and further 2 h at 75 °C. The reaction mixture was poured into water (10 mL) and extracted with diethyl ether (3×10 mL). The combined organic layers were dried over $MgSO_4$ and concentrated under reduced pressure. The product was purified by silica column chromatography (hexane/ethyl acetate 1:1) to give 0.11 g (40%) **5** as pale yellow oil. $[\alpha]_D^{20}$ +76.7 ($CHCl_3$); $\nu_{max}$ (liquid film) 2955, 2869, 2213, 1623, 1467, 1382, 1025 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 6.53 (dd, *J* 7.0, 3.5 Hz, 1H, 5-H), 2.33–2.15 (m, 3H, 3a-H, 6-H), 2.07–1.95 (m, 2H, 3-H, 7-H), 1.87 (m, 1H, 2-H), 1.55–1.08 (m, 11H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H, 5′-H), 1.03 (m, 1H, 2′-H), 0.93 (d, *J* 6.7 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.7 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.5 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 144.8 (C-5), 118.6 (CN), 114.0 (C-4), 54.3 (C-1), 47.7 (C-3a), 41.8 (C-7a), 39.4 (C-4′), 36.0 (C-2′), 35.9 (C-1′), 35.1 (C-7), 28.0 (C-3), 27.9 (C-5′), 25.5 (C-6), 23.8 (C-3′), 23.4 (C-2), 22.8, 22.5 (5′-$CH_3$ and C-6′), 18.7 (1′-$CH_3$), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 274 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 273.24695. $C_{19}H_{31}N$ requires 273.2456.

*4.1.2. (E/Z)-3-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-acrylonitrile (**6**).* Palladium(II) acetate (1.60 mg; 7.1 μmol), tri-*o*-tolylphosphine (4.3 mg; 14.0 μmol), and acrylonitrile (33.0 μL; 0.50 mmol) were added to a solution of the vinyl triflate **4**[23] (0.20 g; 0.50 mmol), acetonitrile (1.0 mL), and tributylamine (1.70 mL). The reaction was performed in a sealed vessel in a single-mode microwave reactor at a maximum output of 150 W, a maximum temperature of 150 °C, and a reaction time of 10 min. The resulting mixture was diluted with diethyl ether (20 mL) and washed with water (2×20 mL) and brine (2×20 mL). The organic layer was dried with $MgSO_4$ and concentrated under reduced pressure. The product was purified by silica column chromatography (hexane/ethyl acetate 10:0→9:1) to give 80 mg (54%; *E*/*Z*-ratio 1:1) **6** as yellow oil. $\nu_{max}$ (liquid film) 2954, 2213, 1608, 1467, 1381, 1218, 965 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 6.89 (d, *J* 16.7 Hz, 0.5×1H, 3″-H), 6.59 (d, *J* 11.6 Hz, 0.5×1H, 3″-H), 6.47 (dd, *J* 6.6, 3.5 Hz, 0.5×1H, 5-H), 6.05 (dd, *J* 6.7, 3.1 Hz, 0.5×1H, 5-H), 5.36 (d, *J* 16.7 Hz, 0.5×1H, 2″-H), 5.14 (d, *J* 11.6 Hz, 0.5×1H, 2″-H), 2.33–2.21 (m, 3H, 3a-H, 6-H), 2.07–1.89 (m, 3H, 2-H, 3-H, 7-H), 1.56–1.09 (m, 11H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H, 5′-H), 1.03 (m, 1H, 2′-H), 0.95 (d, *J* 6.6 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.69 (s, 0.5×3H, 7a-$CH_3$), 0.66 (s, 0.5×3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 153.3, 149.5 (C-3″), 137.4, 131.3 (C-4), 134.6 (C-4), 126.4 (C-5), 118.5 (C-1″), 94.0, 93.7 (C-2″), 54.3, 53.5 (C-1), 49.9, 48.8 (C-3a), 43.0, 42.3 (C-7a), 39.5 (C-4′), 36.1 (C-1′), 35.6 (C-2′), 35.2 (C-7), 28.7, 28.3 (C-5′), 28.0 (C-3), 25.5, 25.2 (C-6), 23.9 (C-2), 23.8 (C-3′), 22.8, 22.6 (5′-$CH_3$ and C-6′), 19.4 ,18.7 (1′-$CH_3$), 11.2, 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 300 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 299.2614. $C_{21}H_{33}N$ requires: 299.2613.

*4.1.3. (E)-4-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-enenitrile (**7**).* Palladium(II) acetate (1.60 mg; 7.1 μmol), tri-*o*-tolylphosphine (4.3 mg; 14.0 μmol), and but-3-enenitrile (40.0 μL; 0.50 mmol) were added to a solution of the vinyl triflate **4** (0.20 g; 0.50 mmol), acetonitrile (1.0 mL), and tributylamine (1.70 mL). The reaction was performed in a sealed vessel in a single-mode microwave reactor at a maximum output of 150 W, a maximum temperature of 150 °C, and a reaction time of 10 min. The resulting mixture was diluted with diethyl ether (20 mL) and washed with water (2×20 mL) and brine (2×20 mL). The organic layer was dried with $MgSO_4$ and concentrated under reduced pressure. The product was purified by silica column chromatography (hexane/ethyl acetate 10:0→9:1) to give 70 mg (44%) **7** as yellow oil. $[\alpha]_D^{20}$ +80.9 ($CHCl_3$); $\nu_{max}$ (liquid film) 2954, 2214, 1725, 1631, 1466, 1380, 1169, 968 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 6.18 (d, *J* 15.8 Hz, 1H, 4″-H), 5.69 (d, *J* 3.0 Hz, 1H, 5-H), 5.52 (dt, *J* 15.8, 5.8 Hz, 1H, 3″-H), 3.12 (d, *J* 5.8 Hz, 2H, 2″-H), 2.26–2.15 (m, 3H, 3a-H, 6-H), 2.05–1.86 (m, 3H, 2-H, 3-H, 7-H), 1.58–1.08 (m, 11H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H, 5′-H), 1.03 (m, 1H, 2′-H), 0.95 (d, *J* 6.5 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.7 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.7 Hz, 3H, 6′-H), 0.67 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 136.9 (C-4″), 136.5 (C-4), 127.6 (C-5), 117.8 (C-1″), 113.9 (C-3″), 53.9 (C-1), 49.7 (C-3a), 42.7 (C-7a), 39.5 (C-4′), 36.1 (C-1′), 36.0 (C-2′), 35.7 (C-7), 28.6 (C-5′), 28.0 (C-3), 24.9 (C-6), 24.1 (C-2), 23.8 (C-3′), 22.8, 22.6 (5′-$CH_3$ and C-6′), 20.9 (C-2″), 18.8 (1′-$CH_3$), 11.2 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 314 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 313.2809. $C_{22}H_{35}N$ requires 313.2769.

*4.1.4. Methanesulfonic acid 4-[(1R,3aR,7aR)-1-((R)-1,5-dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-ynyl ester (**9**).* A solution of alcohol **8**[15] (0.32 g, 1.00 mmol) and dichloromethane (5 mL) was stirred at −10 °C and triethylamine (0.1 mL, 1.20 mmol) was added. Mesyl chloride (0.10 mL, 1.20 mmol) was added dropwise and the reaction mixture was stirred for further 3 h at −10 °C. It was allowed to warm to rt and then washed with a saturated sodium hydrogencarbonate solution (10 mL). The organic layer was dried over $Na_2SO_4$ and concentrated under reduced pressure. The product was purified by silica column chromatography (hexane/ethyl acetate 1:1) to give 0.10 g (27%) **9** as pale yellow oil. $\delta_H$ (500 MHz, $CDCl_3$) 5.92 (dd, *J* 6.9, 3.5 Hz, 1H, 5-H), 4.29 (t, *J* 6.7 Hz, 2H, 1″-H), 3.05 (s, 3H, *S*-$CH_3$), 2.77 (t, *J* 6.8 Hz, 2H, 2″-H), 2.20–2.09 (m, 3H, 3a-H, 6-H), 2.01–1.88 (m, 2H, 3-H, 7-H), 1.76 (m, 1H, 2-H), 1.52 (m, 1H, 5′-H), 1.44–1.08 (m, 10H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H), 1.00 (m, 1H, 2′-H), 0.92 (d, *J* 6.7 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.7 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.7 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (125 MHz, $CDCl_3$) 133.8 (C-5), 121.9 (C-4), 83.0 (C-3″), 81.9 (C-4″), 67.7 (C-1″), 54.7 (C-1), 49.9 (C-3a), 41.8 (C-7a), 39.5 (C-4′), 37.7 (*S*-$CH_3$), 36.2 (C-1′), 36.1 (C-2′), 35.8 (C-7), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.0 (C-2), 23.8 (C-3′), 22.8, 22.5 (5′-$CH_3$ and C-6′), 20.6 (C-2″), 18.7 (1′-$CH_3$), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 395 (83) $[M+H]^+$, 299 (100); (EI, 70 eV): *m*/*z* (%): 394 (51) $[M^+]$, 377 (100).

*4.1.5. 5-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-pent-4-ynenitrile (**10**).* A solution of **9** (0.18 g; 0.46 mmol), sodium cyanide (0.11 g; 2.20 mmol), and DMF (2 mL) was stirred at rt for 2 h. The reaction mixture was concentrated under reduced pressure. The product was purified by silica column chromatography (hexane/ethyl acetate 1:1) to give 0.03 g (21%) **10** as pale yellow oil. $\delta_H$ (400 MHz, $CDCl_3$) 5.94 (dd, *J* 6.7, 3.6 Hz, 1H, 5-H), 2.69–2.54 (m, 4H, 2″-H, 3″-H), 2.21–2.09 (m, 3H, 3a-H, 6-H), 2.07–1.87 (m, 2H, 3-H, 7-H), 1.76

(m, 1H, 2-H), 1.53 (m, 1H, 5′-H), 1.45–0.97 (m, 11H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H), 0.93 (d, *J* 6.6 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 134.0 (C-5), 121.8 (C-4), 118.1 (C-1″), 83.3 (C-4″), 81.8 (C-5″), 54.7 (C-1), 49.9 (C-3a), 41.8 (C-7a), 39.5 (C-4′), 36.2 (C-1′), 36.1 (C-2′), 35.9 (C-7), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.4 (C-2), 23.8 (C-3′), 22.8, 22.5 (5′-$CH_3$ and C-6′), 18.7 (1′-$CH_3$), 17.5 (C-2″), 16.2 (C-3″), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 326 (100) $[M+H]^+$; HRMS (EI, 70 eV) $M^+$, found 325.2760. $C_{23}H_{35}N$ requires 325.2769.

*4.1.6. (3R,3aR,5aS,6R,7R,9bR)-3-((R)-1,5-Dimethylhexyl)-3a-methyl-2,3,3a,4,5,5a,6,7,8,9b-decahydro-1H-cyclopenta[a]naphthalene-6,7-dicarbonitrile (**12a**) and (3R,3aR,5aS,6S,7S,9bR)-3-((R)-1,5-dimethylhexyl)-3a-methyl-2,3,3a,4,5,5a,6,7,8,9b-decahydro-1H-cyclopenta[a]naphthalene-6,7-dicarbonitrile (**12b**).* Diene **11**[23] (0.20 g, 0.73 mmol) and fumaronitrile (0.06 g, 0.73 mmol) were dissolved in toluene (2.2 mL) and heated under reflux at 120 °C for 12 h. The reaction mixture was concentrated under reduced pressure. The product was purified by silica column chromatography (hexane/ethyl acetate 4:1) to give 0.11 g (42%) **12a** as white solid and 0.06 g (23%) **12b** as white solid. Compound **12a**: mp 187–188 °C; $[\alpha]_D^{20}$ +121.8 ($CHCl_3$); $\nu_{max}$ (KBr) 2948, 2244, 1673, 1467, 1382, 1152, 1034, 846 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 5.29 (m, 1H, 9-H), 3.36 (dd, *J* 7.1, 3.3 Hz, 1H, 7-H), 3.13 (dd, *J* 4.2, 3.3 Hz, 1H, 6-H), 2.98 (m, 1H, 5a-H), 2.76 (m, 1H, 8-H), 2.53 (m, 1H, 8-H), 2.45 (m, 1H, 9b-H), 1.99–1.91 (m, 2H, 4-H, 2-H), 1.84–1.76 (m, 2H, 1-H, 5-H), 1.72 (m, 1H, 4-H), 1.59 (m, 1H, 5-H), 1.51 (m, 1H, 5′-H), 1.40–1.26 (m, 6H, 1-H, 2-H, 3-H, 1′-H, 2′-H, 3′-H), 1.20–1.05 (m, 3H, 3′-H, 2×4′-H), 0.99 (m, 1H, 2′-H), 0.90 (d, *J* 5.7 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.76 (s, 3H, 3a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 139.4 (C-9a), 119.7 (6-CN), 117.0 (7-CN), 115.1 (C-9), 57.4 (C-3), 49.1 (C-9b), 41.4 (C-3a), 39.4 (C-4′), 36.6 (C-4), 35.9 (C-1′), 35.8 (C-2′), 32.8 (C-5a), 32.4 (C-6), 28.6 (C-2), 28.0 (C-5′), 27.7 (C-7), 26.0 (C-8), 25.0 (C-5), 23.9 (C-3′), 23.1 (C-1), 22.8, 22.6 (5′-$CH_3$ and C-6′), 18.7 (3a-$CH_3$), 18.4 (1′-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 353 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 352.2921. $C_{24}H_{36}N_2$ requires 352.2878. Compound **12b**: mp 114–115 °C; $[\alpha]_D^{20}$ +9.4 ($CHCl_3$); $\nu_{max}$ (KBr) 2954, 2248, 1670, 1468, 1376, 1140, 1028, 851 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 5.22 (m, 1H, 9-H), 3.11 (dt, *J* 11.0, 6.6 Hz, 1H, 7-H), 2.68 (m, 1H, 8-H), 2.59 (t, *J* 11.0 Hz, 1H, 6-H), 2.55 (m, 1H, 5a-H), 2.45 (m, 1H, 8-H), 2.35 (m, 1H, 9b-H), 2.01 (ddd, *J* 12.9, 8.2, 3.5 Hz, 1H, 5-H), 1.94 (m, 1H, 2-H), 1.90 (dd, *J* 14.2, 8.2 Hz, 1H, 4-H), 1.76 (m, 1H, 1-H), 1.69 (m, 1H, 4-H), 1.51 (m, 1H, 5′-H), 1.45(m, 1H, 5-H), 1.40–1.21 (m, 6H, 1-H, 2-H, 3-H, 1′-H, 2′-H, 3′-H), 1.20–1.04 (m, 3H, 3′-H, 2×4′-H), 0.98 (m, 1H, 2′-H), 0.89 (d, *J* 6.0 Hz, 3H, 1′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.85 (d, *J* 6.6 Hz, 3H, 6′-H), 0.71 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 140.3 (C-9a), 119.2 (6-CN), 118.8 (7-CN), 115.9 (C-9), 57.4 (C-3), 49.0 (C-9b), 41.4 (C-3a), 39.4 (C-4′), 37.8 (C-6), 36.6 (C-4), 36.0 (C-1′), 35.7 (C-2′), 35.4 (C-5a), 29.9 (C-7), 28.6 (C-2, C-8), 28.0 (C-5′), 25.8 (C-5), 23.9 (C-3′), 23.2 (C-1), 22.8, 22.5 (5′-$CH_3$ and C-6′), 18.5 (3a-$CH_3$), 18.3 (1′-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 353 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 352.2908. $C_{24}H_{36}N_2$ requires 352.2878.

*4.1.7. Thioacetic acid S-{4-[(1R,3aR,7aR)-1-((R)-1,5-dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-ynyl}ester (**13**).* Diisopropyl azodicarboxylate (1.01 mL, 5.00 mmol) was added dropwise to a solution of triphenylphosphine (1.31 g, 5.00 mmol) and THF (11.5 mL). The reaction mixture was stirred for 30 min at 0 °C. A solution of alcohol **8** (0.80 g, 2.53 mmol) and thioacetic acid (0.36 mL, 5.00 mmol) in THF (2.67 mL) was added slowly. The mixture was stirred for 1 h at 0 °C and then it was allowed to warm to rt over a period of 1 h. The reaction mixture was concentrated under reduce pressure. The product was purified by silica column chromatography (dichloromethane/hexane 1:1) to give 0.91 g (96%) **13** as pale yellow oil. $[\alpha]_D^{20}$ +10.61 ($CHCl_3$); $\nu_{max}$ (liquid film) 2953, 1695, 1466, 1365, 1132, 946 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 5.90 (dd, *J* 6.5, 3.1 Hz, 1H, 5-H), 3.03 (t, *J* 7.0 Hz, 2H, 1″-H), 2.57 (t, *J* 7.0 Hz, 2H, 2″-H), 3.04 (s, 3H, 2‴-H), 2.20–2.10 (m, 3H, 3a-H, 6-H), 1.99–1.87 (m, 2H, 3-H, 7-H), 1.77 (m, 1H, 2-H), 1.51 (m, 1H, 5′-H), 1.44–1.08 (m, 10H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H), 1.00 (m, 1H, 2′-H), 0.93 (d, *J* 6.5 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 195.6 (C-1‴), 133.0 (C-5), 122.2 (C-4), 85.8 (C-3″), 81.9 (C-4″), 54.8 (C-1), 50.1 (C-3a), 41.8 (C-7a), 39.5 (C-4′), 36.2 (C-1′), 36.1 (C-2′), 35.9 (C-7), 30.6 (C-2‴), 28.7 (C-1″), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.1 (C-2), 23.8 (C-3′), 22.8, 22.5 (5′-$CH_3$ and C-6′), 20.4 (C-2″), 18.7 (1′-$CH_3$), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 375 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 374.2653. $C_{24}H_{38}OS$ requires 374.2643.

*4.1.8. 4-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-yne-1-thiol (**14**).* A solution of **13** (0.28 g, 0.75 mmol) in diethyl ether (4.6 mL) was added slowly to a suspension of lithium aluminum hydride (0.11 g, 3.00 mmol) and diethyl ether (2.8 mL). The reaction mixture was stirred 30 min at rt. The excess of lithium aluminum hydride was destroyed carefully by addition of 1 M hydrogen chloride solution. The mixture was extracted with diethyl ether (2×10 mL), the combined organic layers were washed with water (2×20 mL). It was dried over $MgSO_4$ and concentrated under reduced pressure. The raw product (0.23 g, 93%, colorless oil) was used directly for the next reaction step without further purification.

*4.1.9. General procedure for synthesis of unsymmetrical disulfides (**16a**–**f**).* Over a period of 30 min a solution of **14** (0.50, 1.50 mmol) and dichloromethane (3.75 mL) was added to a solution of the corresponding symmetrical disulfide (**15a**–**f**; 1.50 mmol) and dichloromethane (22.5 mL). The reaction mixture was stirred at rt for 12 h. The mixture was extracted with a 5% sodium hydroxide solution (2×30 mL) and water (2×30 mL). The organic layer was dried over $MgSO_4$ and concentrated under reduced pressure.

*4.1.9.1. 1-(4-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-ynyl)-2-ethyldisulfane (**16a**).* Symmetrical disulfide: diethyl disulfide. The product was purified by silica column chromatography (hexane/ethyl acetate 4:1) to give 0.07 g (15%) as pale yellow oil. $[\alpha]_D^{20}$ +13.9 ($CHCl_3$); $\nu_{max}$ (liquid film) 2952, 1699, 1466, 1381, 1253, 1168, 1016 $cm^{-1}$; $\delta_H$ (500 MHz, $CDCl_3$) 5.90 (dd, *J* 6.8, 3.3 Hz, 1H, 5-H), 2.81 (t, *J* 7.2 Hz, 2H, 1″-H), 2.72 (t, *J* 7.3 Hz, 2H, 1‴-H), 2.69 (t, *J* 7.2 Hz, 2H, 2″-H), 2.20–2.10 (m, 3H, 3a-H, 6-H), 2.00–1.88 (m, 2H, 3-H, 7-H), 1.78 (m, 1H, 2-H), 1.52 (m, 1H, 5′-H), 1.45–1.08 (m, 13H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H, 2‴-H), 0.99 (m, 1H, 2′-H), 0.93 (d, *J* 6.6 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.68 (s, 3H, 5′-$CH_3$); $\delta_C$ (125 MHz, $CDCl_3$) 132.9 (C-5), 122.3 (C-4), 85.9 (C-3″), 81.7 (C-4″), 54.7 (C-1), 50.1 (C-3a), 41.8 (C-7a), 39.5 (C-4′), 38.0 (C-1″), 36.2 (C-1′), 36.1 (C-2′), 35.9 (C-7), 32.9 (C-5″), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.1 (C-2), 23.9 (C-3′), 22.8, 22.5 (5′-$CH_3$ and C-6′), 20.1 (C-2″), 18.7 (1′-$CH_3$), 14.4 (C-6″), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 393 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 392.2592. $C_{24}H_{40}S_2$ requires 392.2572.

*4.1.9.2. 1-(4-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-ynyl)-2-isopropyl disulfane (**16b**).* Symmetrical disulfide: diisopropyl disulfide. The product was purified by silica column chromatography (hexane/ethyl acetate 4:1) to give 0.12 g (20%) as pale yellow oil. $[\alpha]_D^{20}$ +15.6 ($CHCl_3$); $\nu_{max}$ (liquid film) 2953, 1667, 1465, 1378, 1153, 1019 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 5.90 (dd, *J* 6.8, 3.3 Hz, 1H, 5-H), 3.06–2.98 (m, 1H, 1‴-H), 2.81 (t, *J* 7.1 Hz, 2H, 1″-H), 2.68 (t, *J* 7.1 Hz, 2H, 2″-H),

2.18–2.11 (m, 3H, 3a-H, 6-H), 1.99–1.87 (m, 2H, 3-H, 7-H), 1.78 (m, 1H, 2-H), 1.52 (m, 1H, 5′-H), 1.44–1.08 (m, 16H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H, 2‴-H, 3‴-H), 1.00 (m, 1H, 2′-H), 0.93 (d, *J* 6.6 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.5 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.5 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 132.9 (C-5), 122.3 (C-4), 86.0 (C-3″), 81.7 (C-4″), 54.7 (C-1), 50.1 (C-3a), 41.7 (C-7a), 41.1 (C-1‴), 39.5 (C-4′), 38.6 (C-1″), 36.2 (C-1′), 36.0 (C-2′), 35.9 (C-7), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.1 (C-2), 23.9 (C-3′), 22.8 (C-2‴, C-3‴), 22.5 (C-6′, 5′-$CH_3$), 20.1 (C-2″), 18.7 (1′-$CH_3$), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 407 (100) $[M+H]^+$; HRMS (EI, 70 eV): $M^+$, found 406.2745. $C_{25}H_{42}S_2$ requires 406.2728.

*4.1.9.3. 2-{4-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-ynyldisulfanyl}-pyridine (**16c**).* Symmetrical disulfide: 2,2′-dithio dipyridine. The product was purified by silica column chromatography (hexane/ethyl acetate 4:1) to give 0.06 g (9%) as pale yellow oil. $[\alpha]_D^{20}$ +41.1 ($CHCl_3$); $\nu_{max}$ (liquid film) 2951, 1574, 1445, 1417, 1378, 1117, 1043, 758 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 8.46 (d, *J* 4.8 Hz, 1H, 6″-H), 7.77 (dt, *J* 8.0, 0.9 Hz, 1H, 4″-H), 7.64 (m, 1H, 5″-H), 7.09 (ddd, *J* 7.5, 4.9, 1.1 Hz, 1H, 3″-H), 5.90 (dd, *J* 6.3, 3.3 Hz, 1H, 5-H), 2.94 (t, *J* 7.4 Hz, 2H, 1″-H), 2.70 (t, *J* 7.4 Hz, 2H, 2″-H), 2.20–2.10 (m, 3H, 3a-H, 6-H), 2.00–1.87 (m, 2H, 3-H, 7-H), 1.77 (m, 1H, 2-H), 1.52 (m, 1H, 5′-H), 1.44–1.08 (m, 10H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H), 1.00 (m, 1H, 2′-H), 0.92 (d, *J* 6.5 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 160.3 (C-2‴), 149.6 (C-6‴), 137.0 (C-4‴), 133.1 (C-5), 122.2 (C-4), 120.6 (C-5‴), 119.6 (C-3‴), 86.5 (C-3″), 82.0 (C-4″), 54.8 (C-1), 50.8 (C-3a), 41.8 (C-7a), 39.5 (C-4′), 38.1 (C-1″), 36.2 (C-1′), 36.1 (C-2′), 35.9 (C-7), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.1 (C-2), 23.9 (C-3′), 22.8, 22.6 (5′-$CH_3$ and C-6′), 20.0 (C-2″), 18.7 (1′-$CH_3$), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 442 (16) $[M+H]^+$, 177 (100); HRMS (EI, 70 eV): $M^+$, found 441.2468. $C_{27}H_{39}NS_2$ requires 441.2524.

*4.1.9.4. 2-{4-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-ynyldisulfanyl}-phenylamine (**16d**).* Symmetrical disulfide: bis-(2-aminophenyl)-disulfide. The product was purified by silica column chromatography (hexane/ethyl acetate 4:1) to give 0.04 g (6%) as pale yellow oil. $[\alpha]_D^{20}$ +24.6 ($CHCl_3$); $\nu_{max}$ (liquid film) 3471, 3064, 2952, 1608, 1477, 1378, 1309, 746 $cm^{-1}$; $\delta_H$ (500 MHz, $CDCl_3$) 7.42 (d, *J* 7.5 Hz, 1H, 6‴-H), 7.17 (t, *J* 7.4 Hz, 1H, 4‴-H), 6.74 (d, *J* 8.0 Hz, 1H, 3‴-H), 6.68 (t, *J* 7.4 Hz, 1H, 5‴-H), 5.90 (dd, *J* 6.3, 3.0 Hz, 1H, 5-H), 4.38 (s, 2H, $NH_2$), 2.86 (t, *J* 7.4 Hz, 2H, 1″-H), 2.74 (t, *J* 7.4 Hz, 2H, 2″-H), 2.19–2.10 (m, 3H, 3a-H, 6-H), 2.00–1.87 (m, 2H, 3-H, 7-H), 1.75 (m, 1H, 2-H), 1.52 (m, 1H, 5′-H), 1.44–1.08 (m, 10H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H), 1.00 (m, 1H, 2′-H), 0.92 (d, *J* 6.5 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.7 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.7 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 148.7 (C-2‴), 136.1 (C-6‴), 133.7 (C-5), 131.8 (C-4‴), 122.9 (C-4), 119.2 (C-1‴), 119.1 (C-5‴), 116.4 (C-3‴), 86.5 (C-3″), 82.5 (C-4″), 55.5 (C-1), 50.8 (C-3a), 42.5 (C-7a), 40.2 (C-4′), 37.7 (C-1″), 36.9 (C-1′), 36.8 (C-2′), 36.6 (C-7), 28.7 (C-5′), 28.6 (C-3), 25.7 (C-6), 24.8 (C-2), 24.6 (C-3′), 23.6, 23.3 (5′-$CH_3$ and C-6′), 20.5 (C-2″), 19.4 (1′-$CH_3$), 11.8 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 456 (14) $[M+H]^+$, 331 (100); HRMS (EI, 70 eV): $M^+$, found 455.2650. $C_{28}H_{41}NS_2$ requires 455.2680.

*4.1.9.5. 2-{2-[4-(1R,3aR,7aR)-2,3,3a,6,7,7a-Hexahydro-7a-methyl-1-((R)-6-methylheptan-2-yl)-1H-inden-4-yl]-but-3-ynyldisulfanyl}-5-nitropyridine (**16e**).* Symmetrical disulfide: 2,2′-dithiobis(5-nitropyridine). The product was purified by silica column chromatography (hexane/ethyl acetate 4:1) to give 0.11 g (15%) as pale yellow oil. $[\alpha]_D^{20}$ +24.6 ($CHCl_3$); $\nu_{max}$ (liquid film) 2952, 1589, 1517, 1436, 1342, 1216, 1097, 854 $cm^{-1}$; $\delta_H$ (500 MHz, $CDCl_3$) 9.27 (d, *J* 2.6 Hz, 1H, 6‴-H), 8.39 (dd, *J* 8.9, 2.6 Hz, 1H, 4‴-H), 8.01 (d, *J* 8.9 Hz, 1H, 3‴-H), 5.91 (dd, *J* 6.8, 3.4 Hz, 1H, 5-H), 2.99 (t, *J* 7.0 Hz, 2H, 1″-H), 2.78 (t, *J* 7.0 Hz, 2H, 2″-H), 2.21–2.10 (m, 3H, 3a-H, 6-H), 2.01–1.88 (m, 2H, 3-H, 7-H), 1.76 (m, 1H, 2-H), 1.52 (m, 1H, 5′-H), 1.45–1.08 (m, 10H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3′-H, 4′-H), 1.00 (m, 1H, 2′-H), 0.92 (d, *J* 6.6 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.69 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 169.0 (C-2‴), 145.0 (C-6‴), 142.0 (C-5‴), 133.5 (C-5), 131.6 (C-4‴), 122.1 (C-4), 119.3 (C-3‴), 84.9 (C-3″), 82.7 (C-4″), 54.8 (C-1), 50.1 (C-3a), 41.8 (C-7a), 39.5 (C-4′), 38.4 (C-1″), 36.2 (C-1′), 36.1 (C-2′), 35.9 (C-7), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.1 (C-2), 23.9 (C-3′), 22.8, 22.6 (5′-$CH_3$ and C-6′), 20.0 (C-2″), 18.7 (1′-$CH_3$), 11.0 (7a-$CH_3$); (EI, 70 eV): *m*/*z* (%): 486 (17) $[M^+]$, 331 (100); HRMS (EI, 70 eV): $M^+$, found 486.2378. $C_{27}H_{38}N_2O_2S_2$ requires 486.2375.

*4.1.9.6. 2-{4-[(1R,3aR,7aR)-1-((R)-1,5-Dimethylhexyl)-7a-methyl-2,3,3a,6,7,7a-hexahydro-1H-inden-4-yl]-but-3-ynyldisulfanyl}-benzothiazole (**16f**).* Symmetrical disulfide: 2,2′-dithiobis(benzothiazole). The product was purified by silica column chromatography (hexane/ethyl acetate 4:1) to give 0.10 g (13%) as pale yellow oil. $[\alpha]_D^{20}$ +40.8 ($CHCl_3$); $\nu_{max}$ (liquid film) 3062, 2952, 1463, 1378, 1236, 1004, 755 $cm^{-1}$; $\delta_H$ (400 MHz, $CDCl_3$) 7.87 (d, *J* 8.2 Hz, 1H, 4‴-H), 7.80 (d, *J* 8.2 Hz, 1H, 7‴-H), 7.44 (t, *J* 8.1 Hz, 1H, 6‴-H), 7.33 (t, *J* 8.0 Hz, 1H, 5‴-H), 5.90 (dd, *J* 6.8, 3.3 Hz, 1H, 5-H), 3.10 (t, *J* 7.2 Hz, 2H, 1″-H), 2.78 (t, *J* 7.2 Hz, 2H, 2″-H), 2.19–2.09 (m, 3H, 3a-H, 6-H), 2.00–1.86 (m, 2H, 3-H, 7-H), 1.77 (m, 1H, 2-H), 1.53 (m, 1H, 5′-H), 1.44–1.08 (m, 10H, 1-H, 2-H, 3-H, 7-H, 1′-H, 2′-H, 3'-H, 4′-H), 0.99 (m, 1H, 2′-H), 0.92 (d, *J* 6.6 Hz, 3H, 1′-$CH_3$), 0.87 (d, *J* 6.6 Hz, 3H, 5′-$CH_3$), 0.86 (d, *J* 6.6 Hz, 3H, 6′-H), 0.68 (s, 3H, 7a-$CH_3$); $\delta_C$ (100 MHz, $CDCl_3$) 172.5 (C-2‴), 155.1 (C-7a‴), 135.9 (C-3a‴), 132.9 (C-5), 126.3 (C-6‴), 124.6 (C-5‴), 122.2 (C-4), 122.1 (C-4‴), 121.1 (C-7‴), 84.9 (C-3″), 82.5 (C-4″), 54.7 (C-1), 50.0 (C-3a), 41.8 (C-7a), 39.5 (C-4′), 38.6 (C-1″), 36.2 (C-1′), 36.1 (C-2′), 35.9 (C-7), 28.0 (C-5′), 27.9 (C-3), 25.0 (C-6), 24.1 (C-2), 23.9 (C-3′), 22.8, 22.6 (5′-$CH_3$ and C-6′), 20.1 (C-2″), 18.7 (1′-$CH_3$), 11.0 (7a-$CH_3$); (CI, $CH_5^+$); *m*/*z* (%): 498 (18) $[M+H]^+$, 168 (100); HRMS (EI, 70 eV): $M^+$, found 497.2290. $C_{29}H_{39}NS_3$ requires 497.2245.

## 4.2. Biological assay

HL-60 cells (DSMZ, German Collection of Microorganisms and Cell Cultures, Braunschweig, Germany) were maintained in RPMI 1640 medium (PAA Laboratories, Cölbe, Germany) containing 10% fetal bovine serum (FBS, PAA Laboratories, Cölbe, Germany) without antibiotics at 37 °C in a humidified atmosphere containing 5% $CO_2$.

*4.2.1. MTT test.* Cytotoxicity of synthesized compounds was determined by the MTT test according to the method of Mosmann.[28] Solutions of the compounds in DMSO (1 μL, concentrations ranging from $10^{-9}$ to $10^{-4}$ mol/L) were incubated with 99 μL of a suspension of HL 60 cells ($9\times10^5$ cells/mL) in RPMI 1640 medium with 10% FBS in 96-well plates for 24 h at 37 °C. Then, 10 μL of MTT solution in PBS (5 mg/mL) was added and the plate was incubated for another 2 h. The cells were quenched with 190 μL DMSO and after 1 h of continuously shaking of the plates a photometric evaluation on an ELISA plate reader MRX II (Dynex Technologies, Denkendorf, Germany; Software: Revelation 4.06) using the wavelength of 550 nm followed. The $IC_{50}$ values were calculated by using Prism 4 (GraphPad Software, La Jolla, USA).

## Supplementary data

Supplementary data associated with this article can be found in the online version, at doi:10.1016/j.tet.2011.11.076. These data include MOL files and InChiKeys of the most important compounds described in this article.

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