Synthesis and Biological Evaluation of Spiro-δ-lactones as Inhibitors of 17β-Hydroxysteroid Dehydrogenase Type 2 (17β-HSD2)

Article

Synthesis and Biological Evaluation of Spiro-δ-lactones as Inhibitors of 17β-Hydroxysteroid Dehydrogenase Type 2 (17β-HSD2)

Running title: Spiro-δ-lactones as Inhibitors of 17β-HSD2

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ABSTRACT

17β-Hydroxysteroid dehydrogenase type 2 (17β-HSD2) catalyzes the oxidation of the potent estradiol (E2) to the less active estrogen estrone (E1). Inhibitors of this enzyme should maintain the local level of E2 in bone tissue when the E2 concentration in the circulation drops and therefore might be useful for the treatment of osteoporosis. In this work, novel non-steroidal spiro-*δ*-lactone compounds designed as 17β-HSD2 inhibitors were synthesized and their physicochemical and biological properties were investigated. These new spiro-*δ*-lactones are not sufficiently stable for further development and show low inhibition of the enzyme.

GRAPHICAL ABSTRACT

Synthesis and Biological Evaluation of Spiro-δ-lactones as Inhibitors of 17β-Hydroxysteroid Dehydrogenase Type 2 (17β-HSD2)

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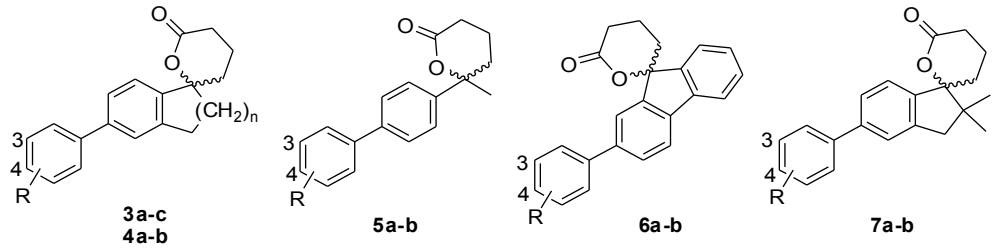


Novel non-steroidal spiro-*δ*-lactones (**3**-**7**), designed as 17β-HSD2 inhibitors were synthesized and their physicochemical and biological properties investigated. These compounds are not sufficiently stable and do not work effectively.

KEYWORDS

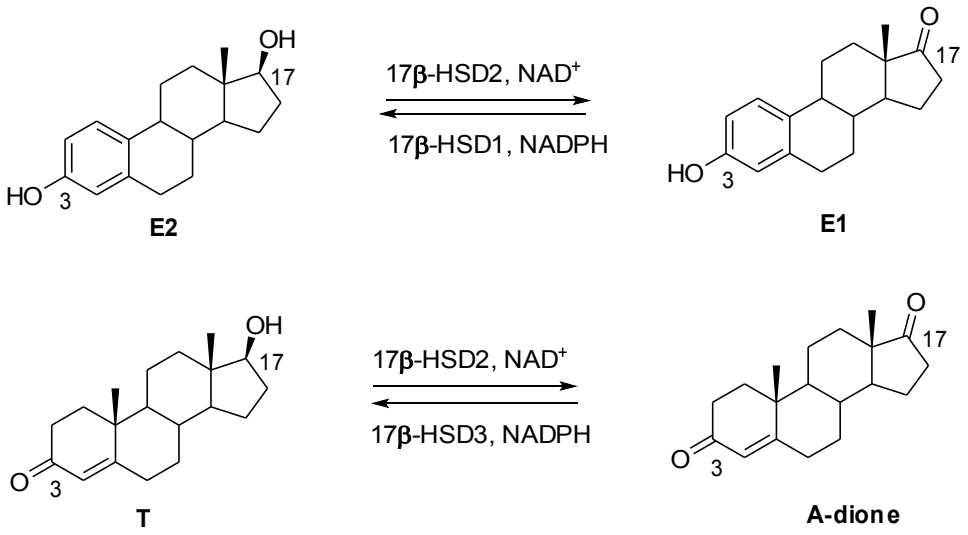
17β-hydroxysteroid dehydrogenase type 2 inhibitors; drug design; osteoporosis; spiro-δ-lactones; steroidomimetics

INTRODUCTION

17β-Hydroxysteroid dehydrogenases (17β-HSDs) [1] are a class of enzymes that catalyze the conversion of 17β-hydroxy estrogens/androgens to 17β-keto analogs and *vice versa*. These enzymes play a key role *in vivo,* controlling the biological activities of the sex hormones in a tissue selective manner [1-5]. Up to now, at least fourteen 17β-HSDs have been identified. All of them belong to the short-chain dehydrogenases/reductases (SDRs) except 17β-HSD5, which is an aldo-keto reductase (AKR) [6].

17β-HSD2 catalyzes the transformation of both estrogenic and androgenic substrates, showing oxidative as well as reductive activity *in vitro*, depending on the presence of the cofactor ($NADP^+$ or NADPH, respectively). However, it has a predominant oxidative activity *in vivo* converting the highly active 17β-hydroxysteroids such as 17β-estradiol (E2) and testosterone (T) to the less active keto analogs estrone (E1) and 4-androstene-3,17-dione (A-dione), respectively, using the cofactor NAD^+ (Fig. 1.). 17β-HSD2 is therefore able to modulate the level in active E2 and T in the target cells. 17β-HSD2 is also able to catalyze the interconversion of 20α-dihydroprogesterone (20α-DHP) to progesterone. This enzyme has not been crystallized yet, no 3D-structure is available.

Fig. 1. 17β-HSD1, 2 and 3 in sex steroid metabolism



Estrogens appear to be essential hormones in bone remodeling: their osteoprotective role is known [7]. Estrogen deficiency or down regulation in the bone cells is believed to be an important risk factor for osteoporosis *e.g.* after menopause or after treatment with aromatase inhibitors [8], which radically prevent the estrogen biosynthesis. Estrogen replacement therapy is efficient against bone loss and osteoporotic fractures [9-11] but it can not be administered to patients because of adverse effects (breast, endometrial and ovarian cancer,

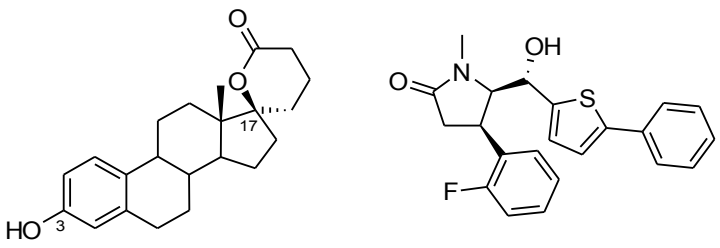
stroke, thromboembolism). There are also substantial evidences that active androgens like T (Fig. 1) and dihydrotestosterone (DHT) may as well be involved in bone formation and may protect the bones against osteoporosis [12, 13]. Controlled increase of active E2 and T in bones of osteoporotic patients will certainly reduce osteoporotic fractures and slow down bone loss by lowering bone resorption and by raising bone formation. Augmentation in E2 and T in bones might be achieved by inhibition of 17β-HSD2 via an intracrine approach [14], where the transformation of E2 and T to inactive precursors will be blocked dominantly in the bone cells.

Thus, inhibition of 17β-HSD2 could help to maintain the local level of E2 and T and therefore provides a novel approach for the treatment of osteoporosis.

As biological counter-parts, 17β-hydroxysteroid dehydrogenase type 1 (17β-HSD1) which oxidizes E1 into E2 (Fig. 1.) and 17β-hydroxysteroid dehydrogenase type 3 (17β-HSD3) which transforms T into A-dione (Fig. 1.), can increase the level of E2 and T, respectively, in the bone cells. These enzymes should not be inhibited [15-16].

Only few potent 17β-HSD2 inhibitors have been reported [1, 17-19]. The group of Poirier described a series of steroidal spirolactone derivatives [20-23], the most potent one is the C17-spiro-δ-lactone **1** (Fig. 2., 65% inhibition at an inhibitor concentration of 1 μM [21]). A novel class of pyrrolidinones [24-26] was also reported as active and selective non-steroidal 17β-HSD2 inhibitors, with **2** (Fig. 2) being the most active compound (IC_{50} = 10 nM). The pyrrolidinone **2** was evaluated in an osteoporosis model using ovariectomized cynomolgus monkeys [27]. A decrease of bone resorption and maintenance of bone formation was observed. Despite high variation and the small effects observed, this *in vivo* experiment validates this approach.

Fig. 2. Described 17β-HSD2 inhibitors



1 **2**

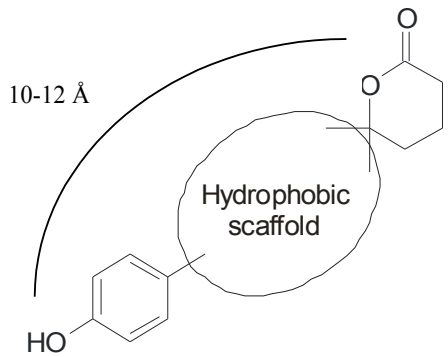
As Bydal [18] describes the spirolactone moiety essential for 17β-HSD2 activity and taking advantage of our experience designing steroidomimetics for different targets (like *e.g.*

CYP19 [28-30], 5α-reductase [31-32], CYP17 [33-34], CYP11B2 [35-39], CYP11B1 [40-41], 17β-HSD1 [42-54] and 17β-HSD2 [19]), the aim of the work described in the following is the design, synthesis and biological evaluation of new non-steroidal spiro-δ-lactone inhibitors of 17β-HSD2.

INHIBITOR DESIGN

In the design process, we decided that steroidal structures should be avoided to limit the side effects due to interaction with steroid hormone receptors. However, the compounds should bind in the substrate binding site of 17β-HSD2 and be capable of mimicking E2 or T: they must have two polar moieties, mimicking positions C3 and C17 of the steroids. In a SAR study [20] based on the steroidal spirolactone **1**, several features were identified, which are important for 17β-HSD2 inhibition. The OH-phenyl moiety is necessary to mimic the 3-OH group of the steroid, protection as methoxy is detrimental for activity. The spirolactone moiety is essential for activity in position C17: a complete loss of activity is observed when the carbonyl group is omitted. The size of the lactone is optimum in case of a δ-lactone (6-membered ring). A hydrophobic scaffold should be present between these two polar groups to mimic the B/C ring of E2 or T. Based on these observations, a pharmacophore model was proposed (Fig. 3.).

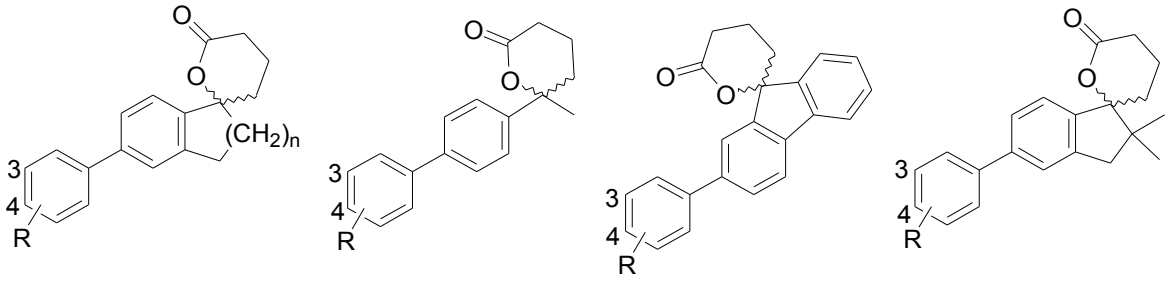
Fig. 3. Simple pharmacophore model



Phenyl indanes and phenyl tetrahydronaphthalenes have often been used as steroidomimetics [33, 35] and will be used as scaffold for the inhibitors. Compounds **3**-**7** (Fig. 4.), bearing the spiro-δ-lactone moiety, combine the features described in the pharmacophore model and a hydrophobic core structure. As the distance between the phenyl-OH and the

lactone should be similar to the corresponding two moieties in E2 (10-12 Å), the hydroxy group on the phenyl of **3-7** should be either in *para* (4) or in *meta* (3) position.

Fig. 4. Designed structures



3a: n = 1, R = 4-OH
3b: n = 1, R = 3-OH
3c: n = 1, R = 4-OMe
4a: n = 2, R = 4-OH
4b: n = 2, R = 3-OH

5a: R = 4-OH
5b: R = 3-OH

6a: R = 4-OH
6b: R = 3-OH

7a: R = 4-OH
7b: R = 3-OH

RESULTS AND DISCUSSION

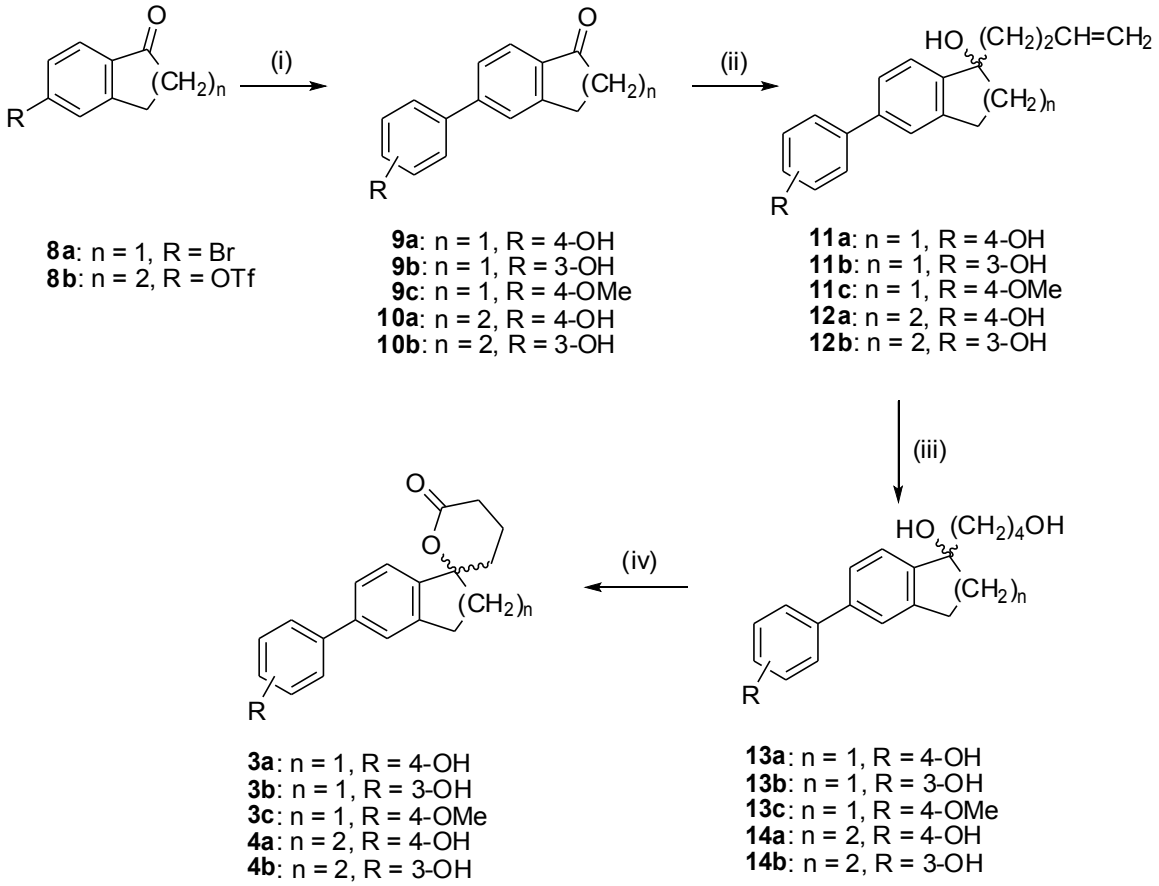
CHEMISTRY

Synthesis of hydroxyphenyl indanes and tetrahydronaphthalenes spirolactone derivatives (3a-c and 4a,b)

Compounds **3a-c** and **4a,b** were prepared in a four steps procedure starting from the 5-bromo-1-indanone (**8a**) and from 6-trifluoromethanesulfonate-1-tetralone (**8b**) (Scheme 1.). First, Suzuki-cross coupling between the bromo (**8a**) or trifluoromethanesulfonate derivatives (**8b**) and the corresponding boronic acids, in presence of a catalytic amount tetrakis triphenylphosphine palladium and two equivalents sodium carbonate 2N afforded **9a-c** and **10a,b** in good yields following the described procedures (Method A) [55, 56]. In a second step, nucleophilic addition of the commercially available Grignard reagent of 4-bromo-1-butene to the carbonyl function of **9a-c** and **10a,b** at 0°C provided the racemic mixture **11a-c, 12a,b** (Method C). Then, the obtained terminal alkene functions were oxidized under hydroboration conditions using BH_3 and subsequent oxidation with Jone's reagent but did not allow isolation of the desired spirolactones. The synthetic pathway could be optimized oxidizing the terminal alkenes with 9-BBN and basic hydrogen peroxide at 0°C (Method D)

and gave the primary alcohols **13a-c** and **14a,b**. In a last step, selective oxidation of the primary hydroxy moiety using Ley's conditions with tetrapropylammonium perruthenate (Method E) [57] followed by spontaneous cyclisation afforded the desired lactones (**3a-c**, **4a,b**).

Scheme 1. Synthesis of compounds **3a-c** and **4a,b**



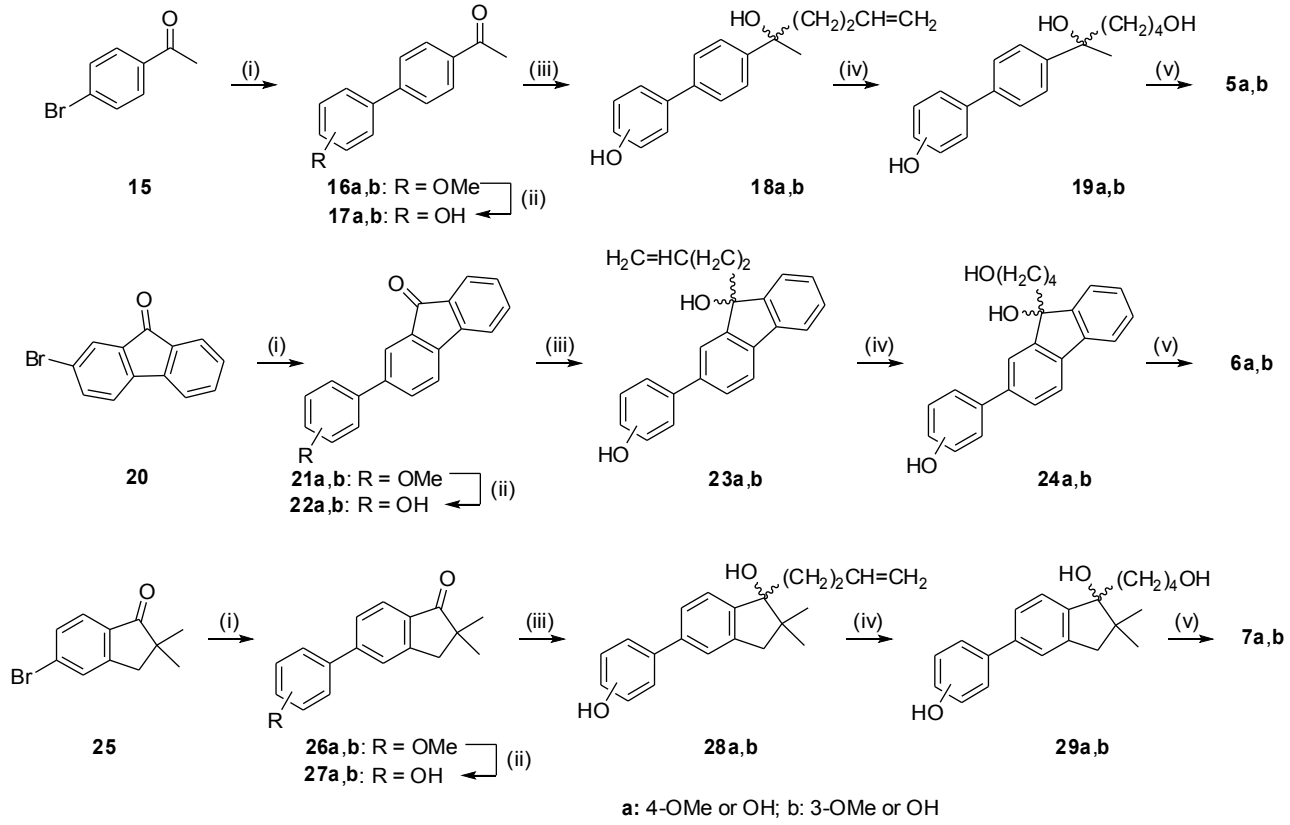
Reagents and conditions: (i): *p*- or *m*-OH(or OMe)PhB$(OH)_2$, $Pd(PPh_3)_4$, 2N Na_2CO_3, PhMe or DME, 80°C, Method A; (ii): CH_2=$CH(CH_2)_2MgBr$, THF, 0°C, Method C; (iii): 9-BBN, 60°C and H_2O, NaOH (3M), H_2O_2, 0°C, Method D; (iv): TPAP, NMO, 4 Å molecular sieve, CH_2Cl_2, rt, Method E.

Synthesis of biphenyl ethyl spirolactone (5a,b), hydroxyphenyl fluorene spirolactone (6a,b) and hydroxyphenyl dimethyl indane spirolactone (7a,b) derivatives

The synthesis of the spirolactones **5-7** is depicted on Scheme 2 and occurred in a five steps reaction. First, methoxylated intermediates **16**, **21** and **26** were obtained by Suzuki-cross coupling reaction of the appropriate brominated ketone (**15**, **20**, **25**) with methoxyphenyl

boronic acids using the standard conditions (catalytic amount of tetrakis triphenylphosphine palladium, 2 equivalents of 2N aqueous sodium carbonate at reflux – method A). Demethylation was performed with boron trifluoride dimethyl sulfide complex (Method B) and led to the hydroxylated ketones **17**, **22** and **27**. Other demethylation reagents like boron tribromide or aluminium chloride were also tried but appeared to be inefficient or led to degradation. The formation of the final spirolactones (**5**-**7**) was achieved following the same synthetic pathway described for **3a**-**c** and **4a**,**b** after nucleophilic addition of the Grignard reagent (Method C), hydroboration-oxydation with 9-BBN and H_2O_2 (Method D) and oxidation with tetrapropylammonium perruthenate (Method E - Scheme 1).

Scheme 2. Synthesis of compounds **5a,b**; **6a,b** and **7a,b**.



Reagents and conditions: (i): *p*- or *m*-MeOPhB(OH)$_2$, 2N Na_2CO_3, $Pd(PPh_3)_4$, PhMe, 80°C, Method A; (ii): $BF_3{\cdot}S(Me)_2$, CH_2Cl_2, rt, Method B; (iii): $CH_2{=}CH(CH_2)_2MgBr$, THF, 0°C, Method C; (iv): 9-BBN, 60°C and H_2O, NaOH (3M), H_2O_2, 0°C, Method D; (v): TPAP, NMO, 4 Å molecular sieve, CH_2Cl_2, rt, Method E.

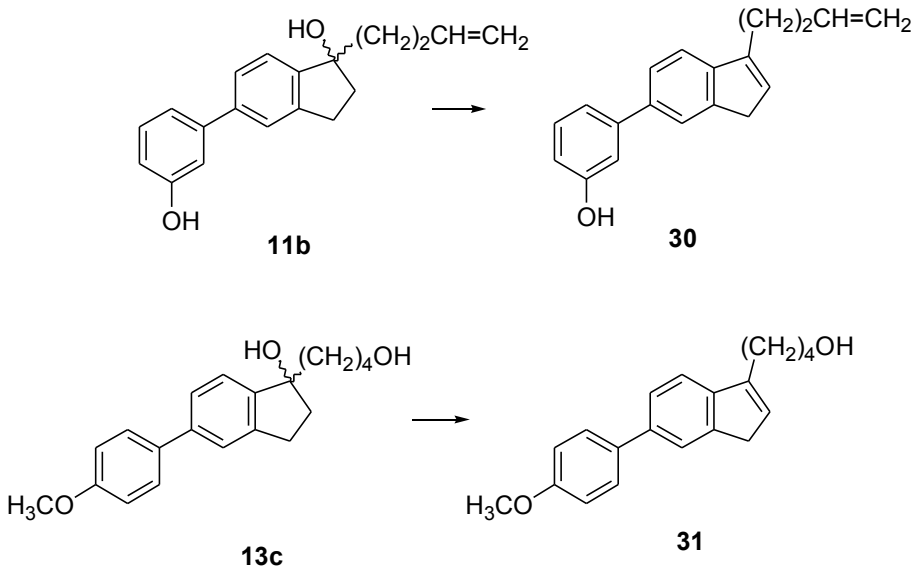
STABILITY STUDY

As the spirolactones **3-7** and precursors were found difficult to isolate with high yields, it was hypothesized that they might have a limited chemical stability. The half-live values of the spirolactones **3a-c**, **4a,b** and **7a,b**, in aqueous buffer solution (20% glycerol, 50 mM KH_2PO_4, 1 mM EDTA disodium salt, pH 7.4) were determined UV-spectrophotometrically and are listed in Table 1. The analyzed compounds are rather unstable and are quickly decomposed at room temperature. The presence of the methoxy on the phenyl increases slightly the stability compared to the hydroxy analog (**3c** *vs* **3a**,**b**).

Derivatives **5a,b**, whose half lives could not be determined by this method, were observed to be rather labile.

We suspect that the spirolactone ring opens and a water molecule is eliminated, as we could isolate and characterize the eliminated products **30** and **31** (Scheme 3). The hydroxy group at 1-position of the indane group is very easily eliminated as the product is conjugated to an aromatic system. The reaction takes place spontaneously at room temperature.

Scheme 3. Spontaneous conversion of compound **11b** and **13c**.



The presence of the adjacent annulated phenyl group is certainly responsible for the instability of the newly synthesized compounds compared to the steroidal analog **1** as the compounds formed like **30** and **31** are thermodynamically more stable.

Compounds **6a,b** and **7a,b**, lacking a proton in α position to the spirolactone ring, were synthesized with the goal to stabilize the compounds and to minimize risk of water elimination. The dimethyl derivatives **7a,b** turned out to be slightly more stable than the

unsubstituted analogs without dimethyl moiety (half-life of **7a**,**b**: 28, 33 min *vs*. **3a**,**b**: 5, 7 min) but still too labile for further drug development.

In opposite, the fluorene spirolactones **6a,b** turned out to present a reasonable chemical stability (half-life > 120 min).

Table 1. Half-life of compounds **3**, **4**, **7**.[a]

Compound	Half-life (min)	Wavelength (nm)
3a	5	255
3b	7	256
3c	57	260
4a	13	257
4b	17	256
7a	28	316
7b	33	316

[a] Determined in aqueous buffer (20% glycerol, 50 mM KH_2PO_4, 1 mM EDTA disodium salt, pH = 7.4).

BIOLOGICAL RESULTS: 17β-HSD2 AND 17β-HSD1 INHIBITORY ACTIVITY

Placental 17β-HSD2 and 17β-HSD1 enzymes were isolated following a described procedure [42, 47]. For determination of 17β-HSD2 inhibition, tritiated E2 was incubated with 17β-HSD2, NAD^+ and the inhibitor (concentration: 1 μM). The amount of formed E1 was quantified by HPLC. Selectivity against 17β-HSD1 was also determined by incubation of tritiated E1, 17β-HSD1, NADH and the inhibitor (concentration: 1 μM). The results are shown in Table 2. None of the synthesized spiro-δ-lactones **3**-**7** has a comparable 17β-HSD2 inhibitory activity as the steroidal derivative **1** (65% inhibition at 1 μM). In case of **3a,b** and **4a,b**, the loss in activity is certainly related to the low stability of the compounds as the duration of the assay is longer (20 min incubation) than the half-life of the compounds (a

short half-life is also expected for **5a,b**). For the spirolactones **6a,b**, which are chemically stable, as well as for **7a,b**, steric hindrance could play a role in the loss in activity as these compounds are much larger than E2. Compound **4b** and **6b** are the two most potent 17β-HSD2 inhibitors identified in this study (25 and 24% inhibition at 1 μM, respectively), with **4b** having the best selectivity profile toward 17β-HSD1.

Table 2. *In vitro* binding potencies in 17β-HSD2 and 17β-HSD1 for the synthesized inhibitors.[a]

Compound	Inhibition of 17β-HSD2 – microsomal fraction (%) at 1μM[b]	Inhibition of 17β-HSD1 – cytosolic fraction (%) at 1μM[c]
1	65	8
3a	0	0
3b	0	0
3c	11	0
4a	0	0
4b	25	8
5a	9	4
5b	4	0
6a	5	6
6b	24	23
7a	14	7

7b	5	5

[a] Mean value of three determinations, standard deviation less than 10%.
[b] Human placental, microsomal fraction, substrate E2, [500 nM], cofactor NAD^+, [1500 μM].
[c] Human placental, cytosolic fraction, substrate E1, [500 nM], cofactor NADH, [500 μM].

EXPERIMENTAL SECTION

CHEMICAL METHODS

Chemical names follow IUPAC nomenclature. Starting materials were purchased from Aldrich, Acros, Lancaster, Roth, Merck or Fluka and were used without purification.

Flash column chromatography (FC) was performed on silica gel (70-200 μm), and reaction progress was monitored by TLC on Alugram SIL G/UV254 (Macherey-Nagel). IR spectra were recorded on a Bruker Vector 33FT-infrared spectrometer (neat sample). Half-lives were measured with a Cary-50 UV/Vis spectrophotometer (Varian, Australia) and all values were measured more than twice and they were repeatable.

^{1}H NMR and ^{13}C NMR spectra were measured on a Bruker AM500 spectrometer (500 MHz) at 300 K. Chemical shifts are reported in δ (parts per million: ppm), by reference to the hydrogenated residues of deuteriated solvent as internal standard ($CDCl_3$: δ 7.26 ppm (^{1}H NMR) and 77 ppm (^{13}C NMR); CD_3COCD_3: 2.05 ppm (^{1}H NMR) and 29.84 and 206.3 ppm (^{13}C NMR); CD_3SOCD_3: 2.50 ppm (^{1}H NMR) and 39.5 ppm (^{13}C NMR). Signals are described as s (singlet), d (doublet), t (triplet), dd (doublet of doublets), ddd (double, double doublet), dt (doublet of triplets), br (broad) and m (multiplet). All coupling constants (*J*) are given in Hertz (Hz).

Mass spectra were measured on a TSQ Quantum (Thermofischer) instrument. No mass peak was detected for the synthesized compounds as the compounds did not ionized in electro-spray mode. All compounds showed more than 95% purity (HPLC determination).

The following compounds were prepared according to previously described procedures: 5-oxo-5,6,7,8-tetrahydronaphthalen-2-yltrifluoromethanesulfonate (**8b**) [56], 5-(4-hydroxyphenyl)-2,3-dihydro-1*H*-inden-1-one (**9a**) [55, 58], 5-(3-hydroxyphenyl)-2,3-dihydro-1*H*-inden-1-one (**9b**) [58], 5-(4-methoxyphenyl)-2,3-dihydro-1*H*-inden-1-one (**9c**) [55], 6-(4-hydroxyphenyl)-3,4-dihydronaphthalen-1(2*H*)-one (**10a**) [55], 6-(3-hydroxyphenyl)-3,4-dihydronaphthalen-1(2*H*)-one (**10b**) [56], 2-bromo-fluoren-9-one (**20)** [59].

General procedure for Suzuki coupling - Method A: A mixture of arylbromide (1 eq), methoxyphenyl boronic acid (1.2 eq), sodium carbonate (2 eq) and tetrakis(triphenylphosphine) palladium (0.1 eq) in an oxygen free toluene/water (1:1) solution was stirred at 80°C for 4-16 h under nitrogen. The reaction mixture was cooled to room temperature. The aqueous layer was extracted with dichloromethane. The combined organic layers were washed with brine, dried over sodium sulfate, filtered and concentrated to dryness.

General procedure for ether cleavage - Method B: To a solution of bis(methoxyphenyl) derivative (1 eq) in dry dichloromethane, borontrifluoride dimethyl sulfide complex (6 eq) was added dropwise at 0°C. The reaction mixture was stirred for 3-14 h at room temperature. Water was added to quench the reaction, and the aqueous layer was extracted with ethyl acetate. The combined organic layers were washed with brine, dried over sodium sulfate, evaporated to dryness under reduced pressure.

General procedure for introducing the but-3-en-1-yl group - Method C: To the Grignard reagent in dry THF prepared from 4-bromobutene (8 eq) and magnesium (6.6 eq) was added dropwise a solution of the ketone derivative (1 eq) in dry THF (5 ml/mmol) and 4-bromobutene (8 eq) under N_2. The reaction mixture was stirred at room temperature for 1 h and the reaction was quenched by addition of 5% $NaHCO_3$. The mixture was extracted with ethyl acetate and the combined organic layers were dried over magnesium sulfate, evaporated to dryness under reduced pressure.

General procedure for hydroboration-oxidation of the terminal alkene - Method D: 9-BBN (6 eq, 0.5 M solution in THF) was added dropwise to a solution of 3-butenyl derivative (1 eq) in THF (3 ml/mmol) under nitrogen at 60°C. The reaction mixture was stirred for 1 h at 60°C and cooled to 0°C. Water (2 ml) was added and the mixture was stirred for 5 min at 0°C. Sodium hydroxide (2 ml/mmol, 3M) was added and after additional 5 min, 2 ml of hydrogen peroxide (30%) was added dropwise. After 30 min the reaction was quenched by addition of $NaHCO_3$ (5% aqueous) and the mixture was extracted with diethyl ether. The organic layers was dried over sodium sulfate and evaporated under reduced pressure.

General procedure for formation of the spirolactones - Method E: To a solution of the diol derivative (1 eq) in anhydrous dichloromethane (4 ml/mmol) was added *N*-

methylmorpholine *N*-oxide (3.9 eq), tetrapropyl ammonium per ruthenate (0.17 eq) and 4 Å molecular sieves (310 mg/mmol) at room temperature. The reaction mixture was stirred for 2-30 min at room temperature, monitored by TLC. The reaction mixture was diluted with dichloromethane (4 ml/mmol) and directly purified.

1-But-3-en-1-yl-5-(4-hydroxyphenyl)indan-1-ol (*RS*, 11a)

Compound **11a** was prepared by reaction of 5-(4-hydroxyphenyl)-2,3-dihydro-1*H*-inden-1-one (**9a,** 70 mg, 0.31 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.25 ml, 2.5 mmol) and magnesium (48 mg, 2.0 mmol) according to the general procedure C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (75 mg, 86%). ^{1}H NMR (CD_3COCD_3) δ 1.77-1.83 (m, 1H), 1.93-1.99 (m, 1H), 2.10-2.20 (m, 2H), 2.22-2.31 (m, 2H), 2.80-2.83 (m, 1H), 2.96-3.02 (m, 1H), 3.92 (br s, 1H), 4.88-4.91 (m, 1H), 5.83-5.91 (m, 1H), 6.89-6.93 (m, 2H), 7.34 (d, *J* = 7.9 Hz, 1H), 7.37-7.42 (m, 2H), 7.46-7.49 (m, 2H), 8.39 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 40.8, 83.0, 114.3, 116.5, 123.4, 124.3, 125.56, 128.9, 133.6, 140.1, 141.6, 144.4, 147.7, 157.8.

1-But-3-en-1-yl-5-(3-hydroxyphenyl)indan-1-ol (*RS*, 11b)

Compound **11b** was prepared by reaction of 5-(3-hydroxyphenyl)-2,3-dihydro-1*H*-inden-1-one (**9b** [44], 60 mg, 0.27 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.22 ml, 2.2 mmol) and magnesium (43 mg, 1.8 mmol) according to the general procedure C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (62 mg, 83%). ^{1}H NMR ($CDCl_3$) δ 1.86-1.92 (m, 1H), 1.95 (br s, 1H), 2.03-2.09 (m, 1H), 2.14-2.21 (m, 2H), 2.23-2.31 (m, 1H), 2.33-2.39 (m, 1H), 2.85-2.91 (m, 1H), 3.03-3.09 (m, 1H), 4.96-4.99 (m, 1H), 5.06 (ddt, *J* = 17.3, 1.9, 1.6 Hz, 1H), 5.36 (br s, 1H), 5.84-5.92 (m, 1H), 6.80 (ddd, *J* = 8.2 Hz, 2.5 Hz, 0.9 Hz, 1H), 6.99 (dd, *J* = 2.2, 1.5 Hz, 1H), 7.13 (ddd, *J* = 7.6 Hz, 1.6 Hz, 0.6 Hz, 1H), 7.28 (t, *J* = 7.9 Hz, 1H), 7.35-7.37 (m, 1H), 7.40-7.43 (m, 2H). ^{13}C NMR ($CDCl_3$) δ 28.7, 29.5, 39.3, 40.1, 83.6, 114.1, 114.2, 114.6, 119.7, 123.1, 123.7, 126.0, 129.9, 138.6, 141.2, 143.0, 143.7, 146.4, 156.0.

1-But-3-en-1-yl-5-(4-methoxyphenyl)indan-1-ol (*RS*, 11c)

Compound **11c** was prepared by reaction of 5-(4-methoxyphenyl)-2,3-dihydro-1*H*-inden-1-one (**9c,** 70 mg, 0.29 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.23 ml, 2.3 mmol) and magnesium (46 mg, 1.9 mmol) according to the general procedure C.

The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 20:1→10:1) as colorless solid (76 mg, 88%). ^{1}H NMR ($CDCl_3$) δ 1.79 (br s, 1H), 1.85-1.91 (m, 1H), 2.02-2.08 (m, 1H), 2.12-2.23 (m, 2H), 2.24-2.31 (m, 1H), 2.33-2.39 (m, 1H), 2.85-2.91 (m, 1H), 3.03-3.09 (m, 1H), 3.85 (s, 3H), 4.96-5.08 (m, 1H), 5.04-5.08 (m, 1H), 5.85-5.93 (m, 1H), 6.95-6.99 (m, 2H), 7.37 (d, *J* = 7.9 Hz, 1H), 7.40-7.44 (m, 2H), 7.51 (d, *J* = 8.8 Hz, 2H). ^{13}C NMR ($CDCl_3$) δ 28.7, 29.5, 39.4, 40.3, 55.3, 83.4, 114.2, 114.5, 123.0, 123.3, 125.6, 128.2, 133.8, 138.7, 141.2, 143.7, 145.8, 159.1.

1-But-3-en-1-yl-6-(4-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol (*RS*, 12a)

Compound **12a** was prepared by reaction of 6-(4-hydroxyphenyl)-3,4-dihydronaphthalen-1(2*H*)-one (**10a** [40], 150 mg, 0.63 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.51 ml, 5.0 mmol) and magnesium (99 mg, 4.1 mmol) according to the general procedure C. The analytically pure product was obtained after purification by FC (dichloromethane/methanol 100:1) as colorless solid (120 mg, 65%). ^{1}H NMR (CD_3COCD_3) δ 1.86-1.90 (m, 4H), 2.08-2.24 (m, 2H), 2.58-2.62 (m, 1H), 2.76-2.83 (m, 2H), 3.04 (t, *J* = 6.1 Hz, 1H), 3.76 (br s, 1H), 4.87-4.91 (m, 1H), 5.00 (ddt, *J* = 17.4 Hz, 3.6 Hz, 1.8 Hz, 1H), 5.81-5.89 (m, 1H), 6.89-6.93 (m, 2H), 6.94-6.97 (m, 1H), 7-25-7.28 (m, 1H), 7.38 (dd, *J* = 8.2 Hz, 2.1 Hz, 1H), 7.46-7.50 (m, 2H), 7.55-7.60 (m, 2H), 8.36 (br s, 1H).

1-But-3-en-1-yl-6-(3-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol (*RS*, 12b)

Compound **12a** was prepared by reaction of 6-(3-hydroxyphenyl)-3,4-dihydronaphthalen-1(2*H*)-one (**10b** [41], 150 mg, 0.63 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.51 ml, 5.0 mmol) and magnesium (99 mg, 4.1 mmol) according to the general procedure C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (98 mg, 53%). ^{1}H NMR ($CDCl_3$) δ 1.80-2.10 (m, 9H), 2.15-2.22 (m, 1H), 2.74-2.87 (m, 2H), 4.94 (ddt, *J* = 10.4 Hz, 1.9 Hz, 1.3 Hz, 1H), 5.02 (ddt, *J* = 17.6 Hz, 1.9 Hz, 1.6 Hz, 1H), 5.64 (br s, 1H), 5.80-5.88 (m, 1H), 6.79 (ddd, *J* = 8.0 Hz, 2.5 Hz, 0.6 Hz, 1H), 7.00 (dd, *J* = 2.5 Hz, 1.6 Hz, 1H), 7.11, (ddd, *J* = 7.6 Hz, 0.9 Hz, 0.6 Hz, 1H), 7.24-7.28 (m, 1H), 7.37 (dd, *J* = 8.2 Hz, 1.9 Hz, 1H), 7.56 (d, *J* = 8.2 Hz, 1H). ^{13}C NMR ($CDCl_3$) δ 19.7, 28.6, 30.0, 36.0, 41.3, 72.6, 114.0, 114.3, 114.5, 119.4, 125.1, 126.7, 127.5, 129.9, 137.1, 138.6, 139.6, 141.1, 142.5, 156.0.

1-(4-Hydroxybutyl)-5-(4-hydroxyphenyl)indan-1-ol (*RS*, 13a)

Compound **13a** was prepared by reaction of 1-but-3-en-1-yl-5-(4-hydroxyphenyl)indan-1-ol **(11a**, 140 mg, 0.5 mmol) with 9-BBN (6 ml, 0.5 M in THF, 3 mmol) and H_2O_2 (1 ml, 30% in H_2O) according to method D. Purification by FC (dichloromethane/methanol 30:1) afforded the desired product as colorless solid (90 mg, 60%). ^{1}H NMR (CD_3COCD_3) δ 1.40-1.47 (m, 1H), 1.50-1.59 (m, 3H), 1.72-1.78 (m, 1H), 1.82-1.92 (m, 1H), 2.08-2.14 (m, 1H), 2.25-2.30 (m, 1H), 2.79-2.83 (m, 1H), 2.95-3.01 (m, 1H), 3.40 (t, *J* = 5.4 Hz, 1H), 3.54 (dt, *J* = 6.3 Hz, 5.3 Hz, 2H), 3.84 (s, 1H), 6.89-6.92 (m, 2H), 7.33 (dd, *J* = 7.6 Hz, 0.9 Hz, 1H), 7.38-7.41 (m, 2H), 7.46-7.49 (m, 2H), 8.38 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.6, 34.4, 40.8, 41.6, 62.5, 83.3, 116.5, 123.4, 124.3, 125.5, 128.8, 133.7, 141.5, 144.5, 148.0, 157.8.

1-(4-Hydroxybutyl)-5-(3-hydroxyphenyl)indan-1-ol (*RS*, 13b)

Compound **13b** was prepared by reaction of 1-but-3-en-1-yl-5-(3-hydroxyphenyl)indan-1-ol (**11b**, 65 mg, 0.23 mmol) with 9-BBN (2.76 ml, 0.5 M in THF, 1.38 mmol) and H_2O_2 (0.45 ml, 30% in H_2O) according to method D. Purification by FC (dichloromethane/methanol 30:1) afforded the desired product as colorless solid (30 mg, 43%). ^{1}H NMR (CD_3COCD_3) δ 1.41-1.48 (m, 1H), 1.50-1.59 (m, 3H), 1.72-1.78 (m, 1H), 1.87-1.93 (m, 1H), 2.09-2.15 (m, 1H), 2.26-2.31 (m, 1H), 2.80-2.87 (m, 1H), 2.96-3.03 (m, 1H), 3.43 (t, *J* = 5.2 Hz, 1H), 3.52-3.57 (m, 2H), 3.90 (s, 1H), 6.82 (ddd, *J* = 8.0 Hz, 2.5 Hz, 0.9 Hz, 1H), 7.08-7.10 (m, 2H), 7.25 (t, *J* = 8.2 Hz, 1H), 7.36 (dd, *J* = 7.4 Hz, 0.9 Hz, 1H), 7.41-7.44 (m, 2H), 8.38 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.6, 30.1, 34.3, 40.7, 41.5, 62.5, 83.3, 114.7, 114.9, 119.0, 123.9, 124.3, 126.0, 130.6, 141.5, 143.8, 144.5, 148.9, 158.7.

1-(4-Hydroxybutyl)-5-(4-methoxyphenyl)indan-1-ol (*RS*, 13c)

Compound **13c** was prepared by reaction of 1-but-3-en-1-yl-5-(4-methoxyphenyl)indan-1-ol (**11c,** 140 mg, 0.48 mmol) with 9-BBN (5 ml, 0.5 M in THF, 2.5 mmol) and H_2O_2 (0.9 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 30:1) afforded the desired product as colorless solid (120 mg, 80%). ^{1}H NMR ($CDCl_3$) δ 1.43-1.51 (m, 1H), 1.56-1.63 (m, 3H), 1.77-1.83 (m, 1H), 1.94-1.99 (m, 1H), 2.10-2.16 (m, 1H), 2.32-2.37 (m, 1H), 2.84-2.90 (m, 1H), 3.02-3.08 (m, 1H), 3.66 (t, *J* = 6.3 Hz, 2H), 3.85 (s, 3H), 6.95-6.98 (m, 2H), 7.35 (d, *J* = 7.9 Hz, 1H), 7.39-7.43 (m, 2H), 7.48-7.52 (m, 2H). ^{13}C NMR ($CDCl_3$) δ 20.5, 29.5, 33.1, 40.0, 40.2, 55.3, 62.7, 83.5, 114.2, 123.0, 123.3, 125.5, 128.2, 133.8, 141.2, 143.7, 146.0, 159.1.

1-(4-Hydroxybutyl)-6-(4-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol (*RS*, 14a)

Compound **14a** was prepared by reaction of 1-but-3-en-1-yl-6-(4-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol (**12a**, 100 mg, 0.34 mmol) with 9-BBN (4 ml, 0.5 M in THF, 2 mmol) and H_2O_2 (0.65 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (71 mg, 67%). ^{1}H NMR (CD_3COCD_3) δ 1.35-1.40 (m, 2H), 1.43-1.56 (m, 3H), 1.79-1.91 (m, 5H), 2.70-2.79 (m, 2H), 3.41 (t, *J* = 5.4 Hz, 1H), 3.51 (dt, *J* = 6.2 Hz, 5.4 Hz, 2H), 3.67 (s, 1H), 6.87 (d, *J* = 8.8 Hz, 2H), 7.22 (s, 1H), 7.34 (dd, *J* = 8.2 Hz, 2.1 Hz, 1H), 7.45 (d, *J* = 8.8 Hz, 2H), 7.55 (d, *J* = 8.2 Hz, 1H), 8.38 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.1, 21.4, 34.4, 36.8, 37.3, 43.6, 62.5, 71.6, 116.5, 124.6, 127.0, 128.0, 128.6, 128.7, 133.2, 137.6, 142.9, 157.8.

1-(4-Hydroxybutyl)-6-(3-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol (*RS*, 14b)

Compound **14b** was prepared by reaction of 1-but-3-en-1-yl-6-(3-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol (**12b**, 100 mg, 0.34 mmol) with 9-BBN (4 ml, 0.5 M in THF, 2 mmol) and H_2O_2 (0.65 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (56 mg, 53%). ^{1}H NMR (CD_3COCD_3) δ 1.32-1.42 (m, 2H), 1.48-1.59 (m, 3H), 1.80-1.86 (m, 5H), 1.90-1.96 (m, 1H), 2.79-2.83 (m, 1H), 3.37 (t, *J* = 5.4 Hz, 1H), 3.52-3.56 (m, 2H), 3.71 (s, 1H), 6.81 (ddd, *J* = 8.2 Hz, 2.2 Hz, 0.9 Hz, 1H), 7.08-7.10 (m, 2H), 7.27-7.29 (m, 1H), 7.28 (s, 1H), 7.40 (dd, *J* = 8.2 Hz, 1.9 Hz, 1H), 7.61 (d, *J* = 8.2 Hz, 1H), 8.33 (s, 1H).). ^{13}C NMR (CD_3COCD_3) δ 21.1, 21.4, 30.7; 34.4, 36.7, 37.4, 43.6, 62.5, 72.1, 114.5, 115.0, 118.9, 125.1, 127.6, 128.0, 130.6, 137.7, 143.4, 143.9, 158.7.

5-(4-Hydroxyphenyl)-2,3,4',5'-tetrahydrospiro[indene-1,2'-pyran]-6'(3'*H*)-one (*RS*, 3a)

Compound **3a** was prepared by reaction of 1-(4-hydroxybutyl)-5-(4-hydroxyphenyl)indan-1-ol (**13a**, 50 mg, 0.17 mmol) with *N*-methylmorpholine *N*-oxide (76 mg, 0.65 mmol) and tetrapropyl ammonium per ruthenate (10 mg, 0.03 mmol) according to method E, Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product as colorless solid (15 mg, 30%). IR (ν, cm^{-1}): 2926, 1694. ^{1}H NMR (CD_3COCD_3) δ 1.98-2.03 (m, 1H), 2.07-2.13 (m, 1H), 2.21-2.26 (m, 1H), 2.38-2.42 (m, 2H), 2.45-2.47 (m, 1H), 2.59-2.62 (m, 2H), 2.91-2.97 (m, 1H), 3.06-3.12 (m, 1H), 6.92 (d, *J* = 8.5 Hz, 2H), 7.41 (d, *J* = 8.5 Hz, 1H), 7.46-7.49 (m, 2H), 7.50 (d, *J* = 8.8 Hz, 2H), 8.42 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 18.1, 32.4, 39.9, 44.3, 50.8, 65.8, 93.1, 116.6, 123.7, 124.5, 126.1, 129.0, 133.2, 142.9, 143.7, 144. 8, 170.4.

5-(3-Hydroxyphenyl)-2,3,4',5'-tetrahydrospiro[indene-1,2'-pyran]-6'(3'*H*)-one (*RS*, 3b)

Compound **3b** was prepared by reaction of 1-(4-hydroxybutyl)-5-(3-hydroxyphenyl)indan-1-ol **(13b**, 20 mg, 0.07 mmol) with *N*-methylmorpholine *N*-oxide (31 mg, 0.26 mmol) and tetrapropyl ammonium per ruthenate (10 mg, 0.01 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product as colorless solid (4 mg, 30%). IR (ν, cm^{-1}): 1696. ^1H NMR ($CDCl_3$) δ 1.91-2.01 (m, 1H), 2.03-2.08 (m, 2H), 2.15-2.20 (m, 1H), 2.31-2.35 (m, 1H), 2.42-2.48 (m, 1H), 2.62-2.76 (m, 2H), 2.87-2.93 (m, 1H), 3.10-3.16 (m, 1H), 5.66 (br s, 1H), 6.83 (ddd, *J* = 8.1 Hz, 2.5 Hz, 0.9 Hz, 1H), 7.03 (dd, *J* = 2.2 Hz, 1.6 Hz, 1H), 7.10 (ddd, *J* = 7.9 Hz, 1.6 Hz, 0.9 Hz, 1H), 7.27 (t, *J* = 7.9 Hz, 1H), 7.32 (dd, *J* = 7.6 Hz, 1.3 Hz, 1H), 7.39-7.43 (m, 2H). ^{13}C NMR ($CDCl_3$) δ 17.2, 29.3, 29.6, 31.8, 39.3, 93.1, 114.3, 114.5, 119.6, 123.5, 123.8, 126.1, 129.9, 142.2, 142.6, 142.7, 143.7, 156.2, 171.5.

5-(4-Methoxyphenyl)-2,3,4',5'-tetrahydrospiro[indene-1,2'-pyran]-6'(3'*H*)-one (*RS*, 3c)

Compound **3c** was prepared by reaction of 1-(4-hydroxybutyl)-5-(4-methoxyphenyl)indan-1-ol (**13c**, 30 mg, 0.10 mmol) with *N*-methylmorpholine *N*-oxide (44 mg, 0.37 mmol) and tetrapropyl ammonium per ruthenate (6 mg, 0.02 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1) afforded the desired product as colorless solid (15 mg, 50%). IR (ν, cm^{-1}): 2932, 1704. ^1H NMR ($CDCl_3$) δ 1.94-2.09 (m, 3H), 2.16-2.22 (m, 1H), 2.32-2.36 (m, 1H), 2.44-2.50 (m, 1H), 2.62-2.71 (m, 2H), 2.89-2.95 (m, 1H), 3.14-3.20 (m, 1H), 3.85 (s, 3H), 6.97 (d, *J* = 8.8 Hz, 2H), 7.35 (d, *J* = 8.0 Hz, 1H), 7.26-7.45 (m, 2H), 7.51 (d, *J* = 8.8 Hz, 2H). ^{13}C NMR ($CDCl_3$) δ 17.3, 29.4, 29.7, 31.9, 39.3, 55.3, 92.9, 114.2, 123.3, 123.5, 125.7, 128.3, 133.5, 142.1, 143.7, 159.3, 170.8.

6-(4-Hydroxyphenyl)-3,4,4',5'-tetrahydro-2*H*-spiro[naphthalene-1,2'-pyran]-6'(3'*H*)-one (*RS*, 4a)

Compound **5a** was prepared by reaction of 1-(4-hydroxybutyl)-6-(4-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol **(14a**, 50 mg, 0.16 mmol) with *N*-methylmorpholine *N*-oxide (73 mg, 0.62 mmol) and tetrapropyl ammonium per ruthenate (10 mg, 0.03 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product (20 mg, 40%) as colorless solid. IR (ν, cm^{-1}): 1696. ^1H NMR

(CD_3COCD_3) δ 1.86-1.98 (m, 4H), 2.07-2.17 (m, 5H), 2.60-2.64 (m, 2H), 2.90-2.94 (m, 1H), 6.90-6.93 (m, 2H), 7.31-7.34 (m, 1H), 7.42-7.46 (m, 2H), 7.48-7.52 (m, 2H), 8.52 (br s, 1H).

6-(3-Hydroxyphenyl)-3,4,4',5'-tetrahydro-2*H*-spiro[naphthalene-1,2'-pyran]-6'(3'*H*)-one (*RS*, 4b)

Compound **4b** was prepared by reaction of 1-(4-hydroxybutyl)-6-(3-hydroxyphenyl)-1,2,3,4-tetrahydronaphthalen-1-ol **(14b**, 26 mg, 0.08 mmol) with *N*-methylmorpholine *N*-oxide (37 mg, 0.31 mmol) and tetrapropyl ammonium per ruthenate (5 mg, 0.01 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product (10 mg, 38%) as colorless solid. IR (ν, cm^{-1}): 1684. ^{1}H NMR (CD_3SOCD_3) δ 1.78-1.86 (m, 3H), 1.93-2.15 (m, 5H), 2.54-2.67 (m, 2H), 2.77-2.89 (m, 2H), 6.76 (ddd, *J* = 8.2 Hz, 2.4 Hz, 0.9 Hz, 1H), 7.00 (t, *J* = 1.9 Hz, 1H), 7.05 (dt, *J* = 8.2 Hz, 0.9 Hz, 1H), 7.24 (t, *J* = 7.9 Hz, 1H), 7.34 (s, 1H), 7.43-7.47 (m, 2H), 9.50 (s, 1H). ^{13}C NMR (CD_3SOCD_3) δ 16.2, 19.2, 28.6, 29.1, 33.1. 34.2, 83.0, 113.3, 114.4, 117.3, 124.3, 126.6, 127.5, 129. 8, 136.9, 138.5, 139.4, 141.1, 157.7, 170.4.

1-(4'-Methoxybiphenyl-4-yl)ethanone (16a)

Compound **16a** was prepared by reaction of 1-(4-bromophenyl)ethanone (**15**, 591 mg, 3.0 mmol) and 4-methoxyphenyl boronic acid (547 mg, 3.6 mmol) with tetrakis(triphenylphosphine)palladium (347 mg, 0.3 mmol) as catalyst according to method A. Purification by FC (hexane/ethyl acetate 10:1) afforded the desired product (650 mg, 96%) as colorless solid. ^{1}H NMR ($CDCl_3$) δ 2.62 (s, 3H), 3.86 (s, 3H), 7.00 (d, *J* = 8.8 Hz, 2H), 7.58 (d, *J* = 8.8 Hz, 2H), 7.64 (d, *J* = 8.5 Hz, 2H), 8.00 (d, *J* = 8.5 Hz, 2H). ^{13}C NMR ($CDCl_3$) δ 26.5, 55.3, 114.3, 126.6, 128.3, 128.8, 132.2, 135.3, 145.3, 159.8, 197.6.

1-(3'-Methoxybiphenyl-4-yl)ethanone (16b)

Compound **16b** was prepared by reaction of 1-(4-bromophenyl)ethanone (**15**, 591 mg, 3.0 mmol) and 3-methoxyphenyl boronic acid (547 mg, 3.6 mmol) with tetrakis(triphenylphosphine)palladium (347 mg, 0.3 mmol) as catalyst according to method A. Purification by FC (hexane/ethyl acetate 10:1) afforded the desired product (655 mg, 97%) as colorless oil. ^{1}H NMR ($CDCl_3$) δ 2.64 (s, 3H), 3.88 (s, 3H), 6.95 (ddd, *J* = 8.2 Hz, 2.5 Hz, 0.6 Hz, 1H), 7.15 (t, *J* = 2.2 Hz, 1H), 7.21 (ddd, *J* = 7.6 Hz, 1.6 Hz, 0.9 Hz, 1H), 7.39 (t, *J* = 8.0 Hz, 1H), 7.66-7.69 (m, 2H), 8.01-8.04 (m, 2H). ^{13}C NMR ($CDCl_3$) δ 26.6, 55.3, 113.1, 113.5, 119.7, 127.3, 128.8, 130.0, 136.0, 141.4, 145.6, 160.0, 197.7.

1-(4'-Hydroxybiphenyl-4-yl)ethanone (17a)

Compound **17a** was prepared by reaction of 1-(4'-methoxybiphenyl-4-yl)ethanone (**16a**, 226 mg, 1 mmol) with borontrifluoride dimethyl sulfide complex (0.63 ml, 6 mmol) according to method B. Purification by FC (hexane/ethyl acetate 10:1) afforded the desired product as colorless solid (170 mg, 80%). ^{1}H NMR (CD_3COCD_3) δ 2.59 (s, 3H), 6.96-6.98 (m, 2H), 7.60-7.62 (m, 2H), 7.71-7.74 (m, 2H), 8.01-8.04 (m, 2H), 8.62 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 26.6, 116.7, 127.0, 129.2, 129.7, 131.7, 136.2, 146.0, 158.8, 197.3.

1-(3'-Hydroxybiphenyl-4-yl)ethanone (17b)

Compound **17b** was prepared by reaction of 1-(3'-methoxybiphenyl-4-yl)ethanone (**16b,** 375 mg, 1.66 mmol) with borontrifluoride dimethyl sulfide complex (1.05 ml, 9.94 mmol) according to method B. Purification by FC (hexane/ethyl acetate 10:1) afforded the desired product as colorless solid (288 mg, 82%). ^{1}H NMR (CD_3COCD_3) δ 2.61 (s, 3H), 6.89-6.92 (m, 1H), 7.17-7.20 (m, 2H), 7.32 (t, J = 8.2 Hz, 1H), 7.75 (d, J = 8.5 Hz, 2H), 8.05 (d, J = 8.5 Hz, 2H), 8.50 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 26.7, 114.8, 116.1, 119.2, 127.8, 129.7, 131.0, 137.1, 142.1, 146.2, 158.9, 197.5.

4'-(1-Hydroxy-1-methylpent-4-en-1-yl)biphenyl-4-ol (*RS*, 18a)

Compound **18a** was prepared by reaction of 1-(4'-hydroxybiphenyl-4-yl)ethanone (**17a**, 132 mg, 0.62 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.51 ml, 4.98 mmol) and magnesium (100 mg, 4.1 mmol) according to method C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (120 mg, 72%). ^{1}H NMR ($CDCl_3$) δ 1.61 (s, 3H), 1.93-2.00 (m, 4H), 2.06-2.10 (m, 1H), 4.93-5.01 (m, 2H), 5.27 (br s, 1H), 5.78-5.82 (m, 1H), 6.90 (d, J = 8.8 Hz, 2H), 7.46-7.49 (m, 4H), 7.52 (d, J = 8.2 Hz, 2H).^{13}C NMR ($CDCl_3$) δ 28.5, 30.3, 43.0, 74.9, 114.6, 115.7, 125.2, 126.4, 128.2, 133.5, 138.7, 139.0, 146.0, 155.2.

4'-(1-Hydroxy-1-methylpent-4-en-1-yl)biphenyl-3-ol (*RS*, 18b)

Compound **18b** was prepared by reaction of 1-(3'-hydroxybiphenyl-4-yl)ethanone (**17b**, 100 mg, 0.47 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.38 ml, 3.77 mmol) and magnesium (74 mg, 3.10 mmol) according to method C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (80 mg, 63%). ^{1}H NMR (CD_3COCD_3) δ 1.57 (s, 3H), 1.86-1.96 (m, 3H), 2.11-2.19 (m, 1H), 3.96 (s, 1H), 4.85 (d, J = 10.4 Hz, 1H), 4.94 (d, J = 17.4 Hz, 1H), 5.76-5.84 (m, 1H),

6.82 (ddd, *J* = 8.0 Hz, 2.2 Hz, 0.9 Hz, 1H), 7.11-7.13 (m, 2H), 7.26 (t, *J* = 8.2 Hz, 1H), 7.57 (s, 4H), 8.37 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 29.2, 30.9, 44.4, 74.0, 114.2 114.5, 115.0, 118.9, 126.4, 127.1, 130.7, 139.5, 140.0, 143.2, 149.0, 158.7.

5-(4'-Hydroxybiphenyl-4-yl)hexane-1,5-diol (*RS*, 19a)

Compound **19a** was prepared by reaction of 4'-(1-hydroxy-1-methylpent-4-en-1-yl)biphenyl-4-ol (**18a**, 110 mg, 0.41 mmol) with 9-BBN (4.92 ml, 0.5 M in THF, 2.46 mmol) and H_2O_2 (0.80 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (73 mg, 62%). ^{1}H NMR (CD_3COCD_3) δ 1.17-1.25 (m, 1H), 1.38-1.48 (m, 3H), 1.53 (s, 3H), 1.77-1.88 (m, 2H), 3.40-3.44 (m, 1H), 3.47-3.50 (m, 2H), 3.89 (s, 1H), 6.92 (d, *J* = 8.8 Hz, 2H), 7.50 (d, *J* = 8.8 Hz, 2H), 7.52 (s, 4H), 8.43 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.3, 30.9, 34.2, 45.2, 62.5, 74.2, 116.5, 126.4, 128.6, 133.1, 139.4, 148.3, 157.8.

5-(3'-Hydroxybiphenyl-4-yl)hexane-1,5-diol (*RS*, 19b)

Compound **19b** was prepared by reaction of 4'-(1-hydroxy-1-methylpent-4-en-1-yl)biphenyl-3-ol (**18b**, 78 mg, 0.29 mmol) with 9-BBN (3.48 ml, 0.5 M in THF, 1.74 mmol) and H_2O_2 (0.70 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (75 mg, 90%). ^{1}H NMR (CD_3COCD_3) δ 1.18-1.25 (m, 1H), 1.41-1.49 (m, 3H), 1.54 (s, 3H), 1.77-1.89 (m, 2H), 3.35 (t, *J* = 5.2 Hz, 1H), 3.46-3.50 (m, 2H), 3.88 (s, 1H), 6.82 (ddd, *J* = 8.2 Hz, 2.5 Hz, 0.9, Hz, 1H), 7.11-7.13 (m, 2H), 7.26 (t, *J* = 8.0 Hz, 1H), 7.55 (s, 4H), 8.38 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.3, 30.9, 34.2, 45.2, 62.5, 74.2, 114.5, 114.9, 118.9, 126.4, 127.0, 130.7, 139.4, 143.3, 149.4, 158.7.

6-(4'-Hydroxybiphenyl-4-yl)-6-methyltetrahydro-2*H*-pyran-2-one (*RS*, 5a)

Compound **5a** was prepared by reaction of 5-(4'-hydroxybiphenyl-4-yl)hexane-1,5-diol (**19a**, 42 mg, 0.15 mmol) with *N*-methylmorpholine *N*-oxide (67 mg, 0.57 mmol) and tetrapropyl ammonium per ruthenate (9 mg, 0.03 mmol) according to method E. Purification by FC (dichloromethane/Ethyl acetate 50:1→30:1) afforded the desired product (12 mg, 28%) as colorless solid. IR (ν, cm^{-1}): 2922, 1695. ^{1}H NMR (CD_3COCD_3) δ 1.56-1.62 (m, 1H), 1.66 (s, 3H), 1.81-1.88 (m, 1H), 2.08-2.12 (m, 1H), 2.35-2.41 (m, 2H), 2.45-2.52 (m, 1H), 6.93 (d, *J* = 8.5 Hz, 2H), 7.43 (d, *J* = 8.5 Hz, 2H), 7.52 (d, *J* = 8.8 Hz, 2H), 7.61 (d, *J* = 8.5 Hz, 2H),

8.50 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 17.6, 31.3, 34.9, 85.4, 116.6, 125.9, 127.2, 128.8, 132.5, 140.6, 144.6, 158.2, 170.9.

6-(3'-Hydroxybiphenyl-4-yl)-6-methyltetrahydro-2*H*-pyran-2-one (*RS*, 5b)

Compound **5b** was prepared by reaction of 5-(3'-hydroxybiphenyl-4-yl)hexane-1,5-diol (**19b**, 50 mg, 0.17 mmol) with *N*-methylmorpholine *N*-oxide (79 mg, 0.67 mmol) and tetrapropyl ammonium per ruthenate (10 mg, 0.03 mmol) according to method E. Purification by FC (CH_2Cl_2/Ethyl acetate 50:1→30:1) afforded the desired product (15 mg, 30%) as colorless solid. IR (ν, cm^{-1}): 2945, 1727. ^{1}H NMR (CD_3COCD_3) δ 1.50-1.64 (m, 1H), 1.67 (s, 3H), 1.82-1.89 (m, 1H), 2.07-2.13 (m, 1H), 2.36-2.42 (m, 2H), 2.45-2.52 (m, 1H), 6.85 (ddd, *J* = 7.9 Hz, 2.2 Hz, 0.9 Hz, 1H), 7.12-7.14 (m, 2H), 7.28 (t, *J* = 8.0 Hz, 1H), 7.47 (dt, *J* = 8.9 Hz, 1.9 Hz, 2H), 7.65 (dt, *J* = 8.8 Hz, 2.2 Hz, 2H), 8.44 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 17.6, 29.8, 31.2, 34.9, 85.4, 114.6, 115.3, 118.94, 125.9, 127.8, 130.8, 140.7, 142.7, 145.6, 158.8, 170.8.

2-(4-Methoxyphenyl)-fluoren-9-one (21a)

Compound **21a** was prepared by reaction of 2-bromo-fluoren-9-one [46] (**20**, 540 mg, 2.08 mmol) and 4-methoxyphenyl boronic acid (380 mg, 2.50 mmol) with tetrakis(triphenylphosphine)palladium (243 mg, 0.21 mmol) as catalyst according to method A. Purification by FC (hexane/dichloromethane 5:1) afforded the desired product (500 mg, 84%) as yellow solid. ^{1}H NMR ($CDCl_3$) δ 3.86 (s, 3H), 6.98 (dt, *J* = 8.8 Hz, 2.2 Hz, 2H), 7.28 (dt, *J* = 7.3 Hz, 1.3 Hz, 1H), 7.48 (dt, *J* = 7.3 Hz, 1.3 Hz, 1H), 7.50-7.53 (m, 2H), 7.55 (dt, *J* = 8.8 Hz, 2.2 Hz, 2H), 7.64-7.67 (m, 2H), 7.85 (d, *J* = 1.6 Hz, 1H). ^{13}C NMR ($CDCl_3$) δ 55.3, 114.3, 120.2, 120.6, 122.4, 124.3, 127.8, 128.8, 132.3, 132.5, 134.4, 134.7, 134.9, 141.9, 142.5, 144.4, 159.6, 193.9.

2-(3-Methoxyphenyl)-fluoren-9-one (21b)

Compound **21b** was prepared from 2-bromo-fluoren-9-one (**20**, 540 mg, 2.08 mmol) and 3-methoxyphenyl boronic acid (380 mg, 2.50 mmol) with tetrakis(triphenylphosphine)palladium (243 mg, 0.21 mmol) as catalyst according to method A. Purification by FC (hexane/dichloromethane 5:1) afforded the desired product (520 mg, 87%) as yellow solid. ^{1}H NMR ($CDCl_3$) δ 3.88 (s, 3H), 6.93 (ddd, *J* = 8.2 Hz, 2.7 Hz, 0.9 Hz, 1H), 7.14, (t, *J* = 2.0 Hz, 1H), 7.20 (ddd, *J* = 7.6 Hz, 1.5 Hz, 0.9 Hz, 1H), 7.30 (dt, *J* = 7.3 Hz, 0.9 Hz, 1H), 7.37 (t, *J* = 8.0 Hz, 1H), 7.49 (dt, *J* = 7.3 Hz, 0.9 Hz, 1H), 7.53-7.57 (m, 2H),

7.64-7.66 (m, 1H), 7.71 (dd, *J* = 7.6 Hz, 1.8 Hz, 1H), 7.89 (d, *J* = 1.5 Hz, 1H). ^{13}C NMR ($CDCl_3$) δ 55.4, 112.3, 113.5, 119.3, 120.4, 120.6, 123.0, 124.4, 129.0, 129.9, 133.2, 134.5, 134.8, 141.3, 142.1, 143.3, 144.2, 160.1, 193.8.

2-(4-Hydroxyphenyl)-fluoren-9-one (22a)

Compound **22a** was prepared by reaction of 2-(4-methoxyphenyl)-fluoren-9-one (**21a**, 286 mg, 1mmol) with borontrifluoride dimethyl sulfide complex (0.63 ml, 6 mmol) according to method B. Purification by FC (dichloromethane/CH_3OH 100:1) afforded the desired product (250 mg, 92%) as yellow solid. ^{1}H NMR (CD_3COCD_3) δ 6.97 (dt, *J* = 8.5 Hz, 2.1 Hz, 2H), 7.37 (dt, *J* = 7.3 Hz, 0.9 Hz, 1H), 7.57-7.63 (m, 4H), 7.73-7.81 (m, 4H), 8.57 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 116.7, 121.6, 122.1, 122.3, 124,6, 128.8, 129.9, 131.8, 133.4, 135.1, 135.6, 135.9, 143.1, 143.2, 145.2, 158.6, 193.8.

2-(3-Hydroxyphenyl)-fluoren-9-one (22b)

Compound **22b** was prepared by reaction of 2-(3-methoxyphenyl)-fluoren-9-one (**21b**, 300 mg, 1.05 mmol) with borontrifluoride dimethyl sulfide complex (0.66 ml, 6.3 mmol) according to method B. Purification by FC (dichloromethane/CH_3OH 100:1) afforded the desired product (260 mg, 91%) as yellow solid. ^{1}H NMR (CD_3COCD_3) δ 6.90 (d, *J* = 7.9 Hz, 1H), 7.17-7.22 (m, 2H), 7.32 (t, *J* = 7.8 Hz, 1H), 7.39 (t, *J* = 7.3 Hz, 1H), 7.59-7.65 (m, 2H), 7.76-7.85 (m, 4H), 8.50 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 114.4, 115.9, 118.8, 121.7, 122.1, 122.9, 124.7, 130.1, 131.0, 134.1, 135.1, 135.6, 136.0, 142.1, 143.1, 144.1, 145.1, 158.9, 193.6.

9-But-3-en-1-yl-2-(4-hydroxyphenyl)-9*H*-fluoren-9-ol (*RS*, 23a)

Compound **23a** was prepared by reaction of 2-(4-hydroxyphenyl)-fluoren-9-one (**22a**, 100 mg, 0.36 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.30 ml, 2.94 mmol) and magnesium (58 mg, 2.43 mmol) according to method C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (50 mg, 41%). ^{1}H NMR (CD_3COCD_3) δ 1.68-1.74 (m, 2H), 2.21-2.27 (m, 2H), 4.56 (s, 1H), 4.76-4.80 (m, 1H), 4.81-4.86 (m, 1H), 5.65-5.72 (m, 1H), 6.95 (dt, *J* = 8.8 Hz, 2.1 Hz, 2H), 7.31 (dt, *J* = 7.3 Hz, 1.2 Hz, 1H), 7.36 (dt, *J* = 7.3 Hz, 1.2 Hz, 1H), 7.53-7.56 (m, 1H), 7.56-7.60 (m, 3H), 7.70-7.73 (m, 1H), 7.74 (dd, *J* = 7.9 Hz, 0.6 Hz, 1H), 7.77 (dd, *J* = 1.5 Hz, 0.6 Hz, 1H), 8.47 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 40.6, 82.5, 114.4, 116.6, 120.6, 121.0,

122.4, 124.5, 127.5, 128.3, 128.8, 129.4, 133.2, 138.9, 139.3, 140.4, 141.5, 150.6, 151.2, 158.1.

9-But-3-en-1-yl-2-(3-hydroxyphenyl)-9*H*-fluoren-9-ol (*RS*, 23b)

Compound **23b** was prepared by reaction of 2-(3-hydroxyphenyl)-fluoren-9-one (**22b**, 136 mg, 0.50 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.41 ml, 4.00 mmol) and magnesium (79 mg, 2.43 mmol) according to method C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (60 mg, 37%). ^{1}H NMR (CD_3COCD_3) δ 1.69-1.75 (m, 2H), 2.21-2.32 (m, 2H), 4.58 (s, 1H), 4.76-4.80 (m, 1H), 4.81-4.86 (m, 1H), 5.65-5.73 (m, 1H), 6.86 (ddd, *J* = 8.2 Hz, 2.5 Hz, 0.9 Hz, 1H), 7.18-7.20 (m, 2H), 7.30 (t, *J* = 8.0 Hz, 1H), 7.33 (dt, *J* = 7.4 Hz, 1.3 Hz, 1H), 7.37 (dt, *J* = 7.4 Hz, 1.3 Hz, 1H), 7.55-7.57 (m, 1H), 7.63 (dd, *J* = 7.9 Hz, 1.9 Hz, 1H), 7.74 (ddd, *J* = 7.3 Hz, 1.3 Hz, 0.6 Hz, 1H), 7.76-7.79 (m, 1H), 7.79 (dd, *J* = 1.6 Hz, 0.6 Hz, 1H), 8.39 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 40.6, 82.5, 114.45, 114.6, 115.2, 119.0, 120.7, 121.0, 123.0, 124.5, 128.1, 128.60, 129.4, 130.8, 139.3, 139.9, 140.2, 141.5, 143.4, 150.8, 151.2, 158.8.

9-(4-Hydroxybutyl)-2-(4-hydroxyphenyl)-9*H*-fluoren-9-ol (*RS*, 24a)

Compound **24a** was prepared by reaction of 9-but-3-en-1-yl-2-(4-hydroxyphenyl)-9*H*-fluoren-9-ol (**23a**, 50 mg, 0.15 mmol) with 9-BBN (1.83 ml, 0.5 M in THF, 0.91 mmol) and H_2O_2 (0.32 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (30 mg, 57%). ^{1}H NMR (CD_3COCD_3) δ 0.99-1.04 (m, 2H), 1.35-1.41 (m, 2H), 2.15-2.25 (m, 2H), 3.33-3.37 (m, 3H), 4.50 (br s, 1H), 6.93-6.97 (m, 2H), 7.29 (dt, *J* = 7.3 Hz, 1.2 Hz, 1 H), 7.34 (dt, *J* = 7.3 Hz, 1.2 Hz, 1H), 7.52-7.55 (m, 1H), 7.56-7.59 (m, 3H), 7.68-7.71 (m, 1H), 7.72 (d, *J* = 7.9 Hz, 1H), 7.75 (d, *J* = 1.5 Hz, 1H), 8.50 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.5, 34.0, 41.3, 62.3, 82.7, 116.6, 120.5, 120.9, 122.3, 124.4, 127.4, 128.3, 128.8, 129.2, 133.3, 139.0, 140.4, 141.4, 150.9, 151.5, 158.0.

9-(4-Hydroxybutyl)-2-(3-hydroxyphenyl)-9*H*-fluoren-9-ol (*RS*, 24b)

Compound **24b** was prepared by reaction of 9-but-3-en-1-yl-2-(3-hydroxyphenyl)-9*H*-fluoren-9-ol (**23b**, 60 mg, 0.18 mmol) with 9-BBN (2.20 ml, 0.5 M in THF, 1.10 mmol) and H_2O_2 (0.36 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (30

mg, 47%). ^{1}H NMR (CD_3COCD_3) δ 0.99-1.06 (m, 2H), 1.35-1.42 (m, 2H), 2.18-2.23 (m, 2H), 3.29-3.39 (m, 2H), 3.29 (t, *J* = 5.3 Hz, 1H), 3.35-3.39 (m, 2H), 4.51 (br s, 1H), 6.85 (ddd, *J* = 8.0 Hz, 2.5 Hz, 1.2 Hz, 1H), 7.17-7.19 (m, 2H), 7.29 (t, *J* = 8.2 Hz, 1H), 7.31 (dd, *J* = 7.3 Hz, 1.2 Hz, 1H), 7.36 (dt, *J* = 7.3 Hz, 1.3 Hz, 1H), 7.52-7.55 (m, 1H), 7.61 (dd, *J* = 7.9 Hz, 1.6 Hz, 1H), 7.68-7.71 (m, 1H), 7.72-7.74 (m, 2H). ^{13}C NMR (CD_3COCD_3) δ 21.5, 34.1, 41.3, 62.2, 82.7, 114.6, 115.2, 118.9, 120.6, 120.9, 122.9, 123.9, 124.5, 128.0, 128.5, 129.3, 130.8, 139.9, 140.3, 141.4, 143.5, 151.1, 151.6, 158.8.

2-(4-Hydroxyphenyl)-4',5'-dihydrospiro[fluorene-9,2'-pyran]-6'(3'*H*)-one (*RS*, 6a)

Compound **6a** was prepared by reaction of 9-(4-hydroxybutyl)-2-(4-hydroxyphenyl)-9*H*-fluoren-9-ol (**24a**, 30 mg, 0.09 mmol) with *N*-methylmorpholine *N*-oxide (40 mg, 0.34 mmol) and tetrapropyl ammonium per ruthenate (5 mg, 0.015 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product (8 mg, 27%) as colorless solid. IR (ν, cm^{-1}): 2923, 1713. ^{1}H NMR (CD_3COCD_3) δ 2.16-2.22 (m, 1H), 2.33-2.40 (m, 3H), 2.88-2.91 (m, 2H), 6.94 (d, *J* = 8.8 Hz, 2H), 7.35 (dt, *J* = 7.6 Hz, 1.3 Hz, 1H), 7.46 (dt, *J* = 7.6 Hz, 1.3 Hz, 1H), 7.58 (d, *J* = 8.7 Hz, 2H), 7.65-7.69 (m, 2H), 7.81-7.83 (m, 2H), 7.86 (dd, *J* = 1.6 Hz, 0.6 Hz, 1H), 8.48 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 18.4, 33.0, 42.8, 90.2, 116.6, 118.6, 121.1, 121.5, 123.0, 125.1, 128.6, 128.7, 129.0, 130.5, 132.8, 138.2, 139.9, 142.1, 148.7, 158.3, 170.5.

2-(3-Hydroxyphenyl)-4',5'-dihydrospiro[fluorene-9,2'-pyran]-6'(3'*H*)-one (*RS*, 6b)

Compound **6b** was prepared by reaction of 9-(4-hydroxybutyl)-2-(3-hydroxyphenyl)-9*H*-fluoren-9-ol (**24b**, 30 mg, 0.09 mmol) with *N*-methylmorpholine *N*-oxide (40 mg, 0.34 mmol) and tetrapropyl ammonium per ruthenate (5 mg, 0.015 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product (10 mg, 30%) as colorless solid. IR (ν, cm^{-1}): 1700. ^{1}H NMR (CD_3COCD_3) δ 2.07-2.13 (m, 2H), 2.17-2.25 (m, 1H), 2.35-2.39 (m, 3H), 6.86 (ddd, *J* = 7.9 Hz, 2.2 Hz, 0.9 Hz, 1H), 7.16-7.19 (m, 2H), 7.29 (dt, *J* = 7.6 Hz, 0.6 Hz, 1H), 7.37, (dt, *J* = 7.6 Hz, 0.9 Hz, 1H), 7.47 (dt, *J* = 7.6 Hz, 1.3 Hz, 1H), 7.68 (dt, *J* = 7.6 Hz, 0.9 Hz, 1H), 7.70 (dd, *J* = 7.9 Hz, 1.6 Hz, 1H), 7.82 (ddd, *J* = 7.6 Hz, 1.3 Hz, 0.6 Hz, 1H), 7.85 (dd, *J* = 7.9 Hz, 0.6 Hz, 1H), 7.88 (dd, *J* = 1.6 Hz, 0.6 Hz, 1H), 8.46 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 18.4, 22.7, 33.0, 42.8, 90.2, 114.8, 115.5, 118.9, 119.1, 121.3, 121.6, 123.6, 125.1, 128.9, 129.3, 130.6, 130.8, 139.2, 139.7, 143.0, 148.1, 148.6, 170.5.

5-(4-Methoxyphenyl)-2,2-dimethylindan-1-one (26a)

Compound **26a** was prepared by reaction of 5-bromo-2,2-dimethyl-2,3-dihydro-1*H*-inden-1-one [45] (**25**, 200 mg, 0.84 mmol) and 4-methoxyphenyl boronic acid (152 mg, 0.88 mmol) with tetrakis(triphenylphosphine)palladium (97 mg, 0.08 mmol) as catalyst according to method A. Purification by FC (hexane/dichloromethane 15:1) afforded the desired product (210 mg, 94%) as colorless solid. ^{1}H NMR ($CDCl_3$) δ 1.26 (s, 6H), 3.03 (s, 2H), 3.87 (s, 3H), 7.00 (d, *J* = 8.8 Hz, 2H), 7.56-7.58 (m, 4H), 7.80 (d, *J* = 7.9 Hz, 1H). ^{13}C NMR ($CDCl_3$) δ 25.4, 42.9, 45.8, 55.4, 114.4, 124.4, 124.8, 126.4, 128.6, 132.7, 133.7, 147.5, 152.9, 160.0. 210.9.

5-(3-Methoxyphenyl)-2,2-dimethylindan-1-one (26b)

Compound **26b** was prepared by reaction of 5-bromo-2,2-dimethyl-2,3-dihydro-1*H*-inden-1-one [45] (**25**, 270 mg, 1.13 mmol) and 3-methoxyphenyl boronic acid (206 mg, 1.36 mmol) with tetrakis(triphenylphosphine)palladium (130 mg, 0.11 mmol) as catalyst according to method A. Purification by FC (hexane/dichloromethane 20:1) afforded the desired product (230 mg, 77%) as colorless solid. ^{1}H NMR ($CDCl_3$) δ 1.25 (s, 6H), 3.03 (s, 2H), 3.86 (s, 3H), 6.96 (ddd, *J* = 8.2 Hz, 2.5 Hz, 0.9 Hz, 1H), 7.15 (dd, *J* = 2.5 Hz, 1.6 Hz, 1H), 7.21 (ddd, *J* = 7.6 Hz, 1.9 Hz, 0.9 Hz, 1H), 7.39 (t, *J* = 8.2 Hz, 1H), 7.58-7.61 (m, 2H), 7.82 (d, *J* = 7.9 Hz, 1H). ^{13}C NMR ($CDCl_3$) δ 25.3, 42.9, 45.8, 55.4, 113.2, 113.7, 119.9, 124.8, 125.1, 127.0, 130.0, 134.4, 141.8, 147.8, 152.8, 160.0, 210.9.

5-(4-Hydroxyphenyl)-2,2-dimethylindan-1-one (27a)

Compound **27a** was prepared by reaction of 5-(4-methoxyphenyl)-2,2-dimethylindan-1-one (**26a**, 105 mg, 0.4 mmol) with borontrifluoride dimethyl sulfide complex (0.25 ml, 2.4 mmol) according to method B. Purification by FC (hexane/ethyl acetate 10:1) afforded the desired product (95 mg, 95%) as colorless solid. ^{1}H NMR (CD_3COCD_3) δ 1.20 (s, 6H), 3.06 (s, 2H), 6.97 (d, *J* = 8.8 Hz, 2H), 7.61 (d, *J* = 8.5 Hz, 2H), 7.64-7.70 (m, 3H), 8.65 (s, 1H).). ^{13}C NMR (CD_3COCD_3) δ 25.5, 43.3, 46.1, 116.8, 125.0, 125.1, 126.8, 129.5, 132.1, 134.3, 148.4, 153.9, 159.0, 210.2.

5-(3-Hydroxyphenyl)-2,2-dimethylindan-1-one (27b)

Compound **27b** was prepared by reaction of 5-(3-methoxyphenyl)-2,2-dimethylindan-1-one (**26b**, 210 mg, 0.8 mmol) with borontrifluoride dimethyl sulfide complex (0.50 ml, 4.74 mmol) according to method B. Purification by FC (hexane/ethyl acetate 8:1) afforded the

desired product (180 mg, 90%) as colorless solid. ^{1}H NMR (CD_3COCD_3) δ 1.20 (s, 6H), 3.08 (s, 2H), 6.91 (ddd, *J* = 7.9 Hz, 2.2 Hz, 0.9 Hz, 1H), 7.17-7.19 (m, 2H), 7.31 (t, *J* = 8.2 Hz, 1H), 7.65-7.67 (m, 1H), 7.71-7.73 (m, 2H), 8.80 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 25.5, 43.2, 46.1, 115.0, 116.2, 119.3, 125.0, 126.0, 127.5, 130.9, 135.1, 142.4, 148.5, 153.9, 158.9, 210.5.

1-But-3-en-1-yl-5-(4-hydroxyphenyl)-2,2-dimethylindan-1-ol (*RS*, 28a)

Compound **28a** was prepared by reaction of 5-(4-hydroxyphenyl)-2,2-dimethylindan-1-one (**27a**, 80 mg, 0.32 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.26 ml, 2.54 mmol) and magnesium (50 mg, 2.09 mmol) according to method C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (75 mg, 77%). ^{1}H NMR (CD_3COCD_3) δ 1.01 (s, 3H), 1.15 (s, 3H), 1.69 (dt, *J* = 12.9 Hz, 4.4 Hz, 1H), 1.79 (ddd, *J* =13.9 Hz, 12.0 Hz, 4.4 Hz, 1H), 2.01-2.09 (m, 1H), 2.37-2.45 (m, 1H), 2.70 (d, *J* = 15.1 Hz, 1H), 2.77 (d, *J* = 15.1 Hz, 1H), 3.65 (d, *J* = 0.6 Hz, 1H), 4.85-4.89 (m, 1H), 4.94-4.9 (m, 1H), 5.79-5.86 (m, 1H), 6.89-6.91 (m, 2H), 7.33 (d, *J* = 7.9 Hz, 1H), 7.34-7.40 (m, 2H), 7.48-7.50 (m, 2H), 8.36 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 22.7, 24.8, 29.3, 36.6, 45.9, 48.6, 84.7, 114.3, 116.5, 123.6, 124.8, 125.7, 128.8, 133.6, 140.4, 141.1, 142.3, 147.0, 157.8.

1-But-3-en-1-yl-5-(3-hydroxyphenyl)-2,2-dimethylindan-1-ol (*RS*, 28b)

Compound **28b** was prepared by reaction of 5-(3-hydroxyphenyl)-2,2-dimethylindan-1-one (**27b**, 170 mg, 0.67 mmol) with a Grignard reagent in THF prepared from 4-bromobutene (0.55 ml, 5.39 mmol) and magnesium (107 mg, 4.45 mmol) according to method C. The analytically pure product was obtained after purification by FC (hexane/ethyl acetate 10:1→6:1) as colorless solid (160 mg, 77%). ^{1}H NMR (CD_3COCD_3) δ 1.01 (s, 3H), 1.16 (s, 3H), 1.69 (dt, *J* = 12.9 Hz, 4.6 Hz, 1H), 1.80 (ddd, *J* = 13.6 Hz, 12.3 Hz, 4.4 Hz, 1H), 2.01-2.09 (m, 1H), 2.38-2.46 (m, 1H), 2.70 (d, *J* = 15.1 Hz, 1H), 2.79 (d, *J* = 15.1 Hz, 1H), 3.69 (d, *J* = 0.6 Hz, 1H), 4.85-4.89 (m, 1H), 4.95-4.99 (m, 1H), 5.78-5.87 (m, 1H), 6.82 (ddd, *J* = 7.8 Hz, 2.2 Hz, 0.9 Hz, 1H), 7.09-7.11 (m, 2H), 7.25 (t, *J* = 8.2 Hz, 1H), 7.35 (d, *J* = 7.9 Hz, 1H), 7.39-7.41 (m, 1H), 7.43 (d, *J* = 7.9 Hz, 1H), 8.35 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 22.6, 24.8, 29.2, 36.6, 45.9, 48.7, 84.7, 114.3, 114.7, 114.9, 119.0, 124.2, 125.3, 125.8, 130.6, 140.4, 141.1, 142.4, 143.8, 148.0, 158.7.

1-(4-Hydroxybutyl)-5-(4-hydroxyphenyl)-2,2-dimethylindan-1-ol (*RS*, 29a)

Compound **29a** was prepared by reaction of 1-but-3-en-1-yl-5-(4-hydroxyphenyl)-2,2-dimethylindan-1-ol (**28a**, 60 mg, 0.19 mmol) with 9-BBN (2.33 ml, 0.5 M in THF, 1.17 mmol) and H_2O_2 (0.38 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (30 mg, 47%). ^{1}H NMR (CD_3COCD_3) δ 1.00 (s, 3H), 1.14 (s, 3H), 1.38-1.55 (m, 3H), 1.65-1.72 (m, 3H), 2.67-2.76 (m, 2H), 3.41 (t, *J* = 5.4 Hz, 1H), 3.50-3.58 (m, 2H), 3.58 (s, 1H), 6.90 (d, *J* = 8.8 Hz, 2H), 7.31-7.37 (m, 3H), 7.47 (d, *J* = 8.8 Hz, 2H), 8.40 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.2, 22.8, 24.9, 34.6, 37.3, 46.0, 48.6, 62.56, 84.9, 116.5, 123.5, 124.8, 125.7, 128.9, 133.7, 140.9, 142.3, 147.4, 157.8.

1-(4-Hydroxybutyl)-5-(3-hydroxyphenyl)-2,2-dimethylindan-1-ol (*RS*, 29b)

Compound **29b** was prepared by reaction of 1-but-3-en-1-yl-5-(3-hydroxyphenyl)-2,2-dimethylindan-1-ol (**28b**, 131 mg, 0.42 mmol) with 9-BBN (5.10 ml, 0.5 M in THF, 2.55 mmol) and H_2O_2 (0.84 ml, 30% in H_2O) according to the method D. Purification by FC (dichloromethane/methanol 50:1→30:1) afforded the desired product as colorless solid (58 mg, 43%). ^{1}H NMR (CD_3COCD_3) δ 1.00 (s, 3H), 1.14 (s, 3H), 1.38-1.57 (m, 3H), 1.65-1.73 (m, 3H), 2.68-2.78 (m, 2H), 3.39 (t, *J* = 5.4 Hz, 1H), 3.50-3.54 (m, 2H), 3.62 (s, 1H), 6.81 (ddd, *J* = 8.2 Hz, 2.2 Hz, 1.3 Hz, 1H), 7.08-7.10 (m, 2H), 7.25 (t, *J* = 8.2 Hz, 1H), 7.35 (d, *J* = 7.9 Hz, 1H), 7.38-7.42 (m, 2H), 8.38 (s, 1H). ^{13}C NMR (CD_3COCD_3) δ 21.2, 22.7, 24.8, 34.6, 37.2, 45.9, 48.6, 62.6, 84.9, 114.6, 114.9, 119.0, 124.0, 125.3, 125.8, 130.6, 141.0, 142.4, 143.8, 148.4, 158.7.

5-(4-Hydroxyphenyl)-2,2-dimethyl-2,3,4',5'-tetrahydrospiro[indene-1,2'-pyran]-6'(3'*H*)-one (*RS*, 7a)

Compound **7a** was prepared by reaction of 1-(4-hydroxybutyl)-5-(4-hydroxyphenyl)-2,2-dimethylindan-1-ol (**29a**, 30 mg, 0.09 mmol) with *N*-methylmorpholine *N*-oxide (42 mg, 0.36 mmol) and tetrapropyl ammonium per ruthenate (5 mg, 0.015 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product (10 mg, 34%) as colorless solid. IR (ν, cm^{-1}): 2923, 1695. ^{1}H NMR (CD_3COCD_3) δ 1.11 (s, 3H), 1.13 (s, 3H), 1.99-2.02 (m, 1H), 2.10-2.14 (m, 3H), 2.50-2.57 (m, 1H), 2.64-2.69 (m, 1H), 2.75 (d, *J* = 15.1 Hz, 1H), 2.95 (d, *J* = 15.4 Hz, 1H), 6.92 (d, *J* = 8.8 Hz, 2H), 7.35-7.44 (m, 3H), 7.50 (d, *J* = 8.8 Hz, 2H), 8.46 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 17.9, 23.1, 23.9, 27.3, 45.9, 49.3, 90.8, 116.6, 124.0, 125.0, 125.4, 128.9, 133.1, 140.4, 143.6, 148.3, 158.7, 170.6.

5-(3-Hydroxyphenyl)-2,2-dimethyl-2,3,4',5'-tetrahydrospiro[indene-1,2'-pyran]-6'(3'*H*)-one (*RS*, 7b)

Compound **7b** was prepared by reaction of 1-(4-hydroxybutyl)-5-(3-hydroxyphenyl)-2,2-dimethylindan-1-ol **(29b**, 40 mg, 0.12 mmol) with *N*-methylmorpholine *N*-oxide (56 mg, 0.48 mmol) and tetrapropyl ammonium per ruthenate (7 mg, 0.02 mmol) according to method E. Purification by FC (dichloromethane/ethyl acetate 50:1→30:1) afforded the desired product (14 mg, 35%) as colorless solid. IR (ν, cm^{-1}): 2924, 1690. ^{1}H NMR (CD_3COCD_3) δ 1.11 (s, 3H), 1.13 (s, 3H), 1.98-2.03 (m, 1H), 2.08-2.14 (m, 3H), 2.51-2.58 (m, 1H), 2.65-2.71 (m, 1H), 2.77 (d, *J* = 15.4 Hz, 1H), 2.96 (d, *J* = 15.4 Hz, 1H), 6.84 (ddd, *J* = 8.2 Hz, 2.5 Hz, 0.9 Hz, 1H), 7.10-7.12 (m, 2H), 7.27 (t, *J* = 8.0 Hz, 1H), 7.43 (d, *J* = 7.9 Hz, 1H), 7.46-7.50 (m, 2H), 8.42 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 17.9, 23.1, 23.8, 27.3, 30.6, 45.9, 49.3, 94.7, 114.8, 115.3, 119.1, 124.6, 125.0, 126.0, 130.7, 142.5, 143.3, 143.6, 144.8, 158.8, 170.6.

3-(3-(But-3-enyl)-1*H*-inden-6-yl)phenol (30)

Compound **30** was obtained after the spontaneous conversion of compound **11b** at room temperature. ^{1}H NMR ($CDCl_3$) δ 2.46-2.51 (m, 2H), 2.66-2.70 (m, 2H), 3.40 (d, *J* = 1.9 Hz, 2H), 4.92 (d, *J* = 3.8 Hz, 1H), 5.03 (dd, *J* = 10.1 Hz, 1.6 Hz, 1H), 5.12 (dd, *J* = 17.0 Hz, 1.6 Hz, 1H), 5.92-6.00 (m, 1H), 6.28 (t, *J* = 1.6 Hz, 1H), 6.81 (ddd, *J* = 7.9 Hz, 2.5 Hz, 0.9 Hz, 1H), 7.10 (t, *J* = 2.0 Hz, 1H), 7.21 (dt, *J* = 7.6 Hz, 1.3 Hz, 1H), 7.31 (t, *J* = 7.9 Hz, 1H), 7.42 (d, *J* = 7.9 Hz, 1H), 7.53 (dd, *J* = 7.9, 1.6 Hz, 1H), 7.67 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 27.2, 32.1, 37.8, 113.7, 114.1, 114.8, 119.0, 119.8, 122.6, 125.2, 128.6, 129.9, 137.3, 138.4, 143.6, 143.7, 144.9, 145.1, 155.8.

4-(6-(4-Methoxyphenyl)-1*H*-inden-3-yl)butan-1-ol (31)

Compound **30** was obtained after the spontaneous conversion of compound **13c** at room temperature. ^{1}H NMR ($CDCl_3$) δ 1.30-1.35 (m, 2H), 1.68-1.73 (m, 2H), 1.77-1.83 (m, 2H), 2.59-2.63 (m, 1H), 3.39 (d, *J* = 1.6 Hz, 2H), 3.71 (t, *J* = 6.5 Hz, 2H), 3.86 (s, 3H), 6.22-6.24 (m, 1H), 6.96-7.00 (m, 2H), 7.40 (d, *J* = 7.9 Hz, 1H), 7.48-7.51 (m, 1H), 7.54-7.57 (m, 2H), 7.65 (br s, 1H). ^{13}C NMR (CD_3COCD_3) δ 24.2, 27.5, 32.7, 37.8, 55.3, 62.9, 114.2, 119.1, 122.3, 124.8, 128.1, 128.7, 134.4, 137.5, 144.0, 144.1, 158.9.

BIOLOGICAL METHODS

[2,4,6,7-^{3}H]-E1 and [2,4,6,7-^{3}H]-E2 were purchased from Perkin Elmer, Boston. Quickszint Flow 302 scintillator fluid was bought from Zinsser Analytic, Frankfurt. Other chemicals were purchased from Sigma, Roth or Merck.

17β-HSD1 and 17β-HSD2 Enzyme preparation from human placental enzyme

17β-HSD1 and 17β-HSD2 were obtained from human placenta according to previously described procedures [47, 60]. Fresh human placenta was homogenized and the enzymes were separated by fractional centrifugation at 1000 g, 10000 g and 150000 g. For the purification of 17β-HSD1, the cytosolic fraction was precipitated with ammonium sulfate. 17β-HSD2 was obtained from the microsomal fraction. Aliquots containing 17β-HSD1 or 17β-HSD2 were stored frozen.

Inhibition of 17β-HSD2 in cell-free assay

Inhibitory activities were evaluated by an established method with minor modifications [22, 47, 60]. Briefly, the enzyme preparation was incubated with NAD^+ [1500 µM] in the presence of potential inhibitors at 37°C in a phosphate buffer (50 mM) supplemented with 20% of glycerol and EDTA 1mM. Inhibitor stock solutions were prepared in DMSO. Final concentration of DMSO was adjusted to 1% in all samples. The enzymatic reaction was started by addition of a mixture of unlabelled- and [^{3}H]- E2 (final concentration: 500 nM, 0.11 µCi). After 20 min, the incubation was stopped with $HgCl_2$ and the mixture was extracted with ether. After evaporation, the steroids were dissolved in acetonitrile. E1 and E2 were separated using acetonitrile/water (45:55) as mobile phase in a C18 RP chromatography column (Nucleodur C18, 3µm, Macherey-Nagel, Düren) connected to a HPLC-system (Agilent 1100 Series, Agilent Technologies, Waldbronn). Detection and quantification of the steroids were performed using a radioflow detector (Berthold Technologies, Bad Wildbad). The conversion rate was calculated according to the following equation: $\%conversion = \frac{\%E1}{\%E1 + \%E2} \bullet 100$. Each value was calculated from at least three independent experiments.

Inhibition of 17β-HSD1 in cell-free assay

The 17β-HSD1 inhibition assay was performed similarly to the 17β-HSD2 test. The microsomal fraction was incubated with NADH [500 µM], test compound and a mixture of unlabelled- and [^{3}H]-E1 (final concentration: 500 nM, 0.15 µCi) for 10 min at 37°C. Further

treatment of the samples and HPLC separation was carried out as mentioned above for 17β-HSD2.

CONCLUSION

In this study, we applied a ligand based approach to design new non-steroidal 17β-HSD2 inhibitors bearing a spiro-δ-lactone moiety, which is believed to be responsible for the 17β-HSD2 inhibitory activity of a steroidal compound (**1**)[20]. Eleven spiro-δ-lactone steroidomimetics were synthesized and tested for 17β-HSD2 inhibitory activity. None of the compounds could reach the activity of the steroidal spiro-δ-lactone **1**, they were either inactive or showed a low activity (25% for **4b** and 24% for **6b**) when tested at a concentration of 1 μM.

Several problems, met during the synthesis, obliged us to investigate a new synthesis pathway for the formation of the spiro-δ-lactone moiety, different to the one described [20] for the synthesis of **1.** The first step of the synthesis (introduction of the alkene chain on the tetralone by nucleophilic addition with a Grignard reagent) was performed as described for **1**. The alkene moiety could be oxidized in primary alcohol by hydroboration as mentionned for **1** but then oxidation of this primary alcohol in carboxylic acid using the Jone's reagent failed: only elimination of water could be observed under these conditions. The oxidation of the primary hydroxy moiety could be achieved under Ley's conditions using tetrapropylammonium perruthenate and afforded, after spontaneous cyclisation, the designed compounds **3**-**7**.

Despite the fact that compound **1** was stable, it appeared that the new derivatives decomposed and were very difficult to purify. This chemical instability was confirmed and quantified by measurement of the half-life in aqueous buffer. It might be due to the fact that the C ring of the steroidal **1** is not aromatic, while the phenyl ring, present next to the spirolactone in the synthesized tetrahydronaphathlene or tetrahydroindane derivatives, favors the elimination of a water molecule. Attempts to hinder this elimination (synthesis of **7a,b**) did not substantially improve the stability.

In case of the stable fluorene derivatives **6b**, only a weak inhibitory activity was monitored (24% inhibition at 1 μM). This can be explained by different hypothesis: a) the spirolactone moiety in **6b** might be in a completely different area compared to the one in **1** and either might not fit there or can not achieve favorable interactions as in **1**; b) the supplementary phenyl ring present in the fluorene moiety might lead to steric hindrance preventing its binding in the active site. No new interaction could be reached by the introduction of the extra

phenyl ring. Due to its stability, **6b** would be the best candidate within this series of derivatives, for structural optimization, trying to increase activity and selectivity toward 17β-HSD1.

As 17β-HSD2 is membrane-bound, it has not been yet crystallized and its 3D-structure remains unknown. In order to better understand the interactions achieved by the spiro-δ-lactone **1** with the enzyme and to simplify the drug design process, it would be very useful to have access to a good and reliable homology model of the enzyme.

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