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PAPER

Aromatic A-ring analogues of orobanchol, new germination stimulants for seeds of parasitic weeds†

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Strigolactones are signaling compounds in plants of increasing importance. In this paper the focus is on their activity as germinating agents for seeds of parasitic weeds. The syntheses of aromatic A-ring analogues of the germination stimulant orobanchol have been described. Starting substrate is the ABC unit of the stimulant GR24. Oxidation at the C-4 position gives a 4-oxo derivative which on subsequent reduction produces two C-4 epimeric alcohols, *syn* and *anti* in a ratio of 82 : 3. For practical access of the C-4 *anti* alcohol, the predominant *syn* epimer is inverted by a Mitsunobu procedure. The *anti* C-4 alcohol is then coupled with the D-ring in a one-pot two-step process involving a formylation and a reaction with bromobutenolide to give a mixture of the diastereomeric aromatic A-ring analogues of orobanchol. In contrast, the *syn* C-4 alcohol cannot be coupled directly with the D-ring. Protection of the C-4 *syn* OH is a prequisite. The best protecting function is the SEM group as deprotection after coupling with the D-ring can then readily be achieved. The structures of these new analogues have been ascertained by X-ray analyses. Both diastereomers of the C-4 *syn* as well as the C-4 *anti* orobanchol analogues have been tested as germination agents of seeds of *Striga hermonthica* and *Orobanche ramosa*. In addition, the acetates of both epimeric C-4 alcohols have been prepared and tested. Both diastereomers of the 4-oxo derivative have been prepared and bioassayed as well. The bioassays reveal that the diastereomers having the natural relative configuration are most active. The data also suggest that hydrogen bonding is not an important factor in the binding of the stimulant molecules in the receptor.

Introduction

Strigolactones constitute an important family of bioactive terpenoids that are present in root exudates of several plants and that were initially identified as germinating agent for seeds of root parasites such as *Striga* and *Orobanche* spp.[1–3] Interestingly, recent findings show that strigolactones also serve as the branching factor for arbuscular mycorrhizal (AM) fungi[4–7] and as inhibitor of shoot branching and bud outgrowth in various plants.[8,9] Currently, strigolactones are considered as a new type of plant hormones.[10] Several recent reviews demonstrate the current interest in these compounds.[2,5,10–12] The isolation of natural strigolactones is extremely difficult due to the very small quantities that are present in root exudates.[1,11] The estimated production is *ca.* 15 pg per day per plant. Strigol (see Fig. 1) was the first natural strigolactone that was isolated in 1966 from cotton roots.[13a] The details of its chemical structure were elucidated many years later.[13b] At present several natural strigolactones are known,[1,3] of which typical examples are depicted in Fig. 1. All strigolactones known thus far consist of an annelated three ring ABC skeleton invariably attached to the same D-ring. The structural differences of strigolactones are mainly present in the AB part of these molecules. The total syntheses of natural strigolactones involve multistep sequences; especially installing the correct stereochemistry has been proven very demanding.[14,15] There has been a continuous quest for structurally simplified strigolactones in which the bioactivity is predominantly retained.[16] The most notable synthetic analogue is GR24 wherein the A-ring of strigolactones is replaced by an aromatic ring.[17] This analogue has a high germination activity towards seeds of parasitic weeds and is widely used as a standard positive control in germination experiments.[16] By systematically simplifying the strigolactone structure the bioactiphore in these biocompounds has been established and found to be residing in the CD-part of these molecules.[16,18] Substituents, especially a hydroxy group, can considerably influence, in most cases enhance,

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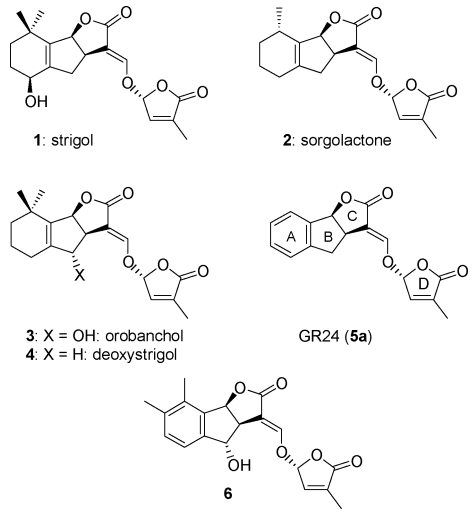


Fig. 1 Natural strigolactones and synthetic analogue GR24.

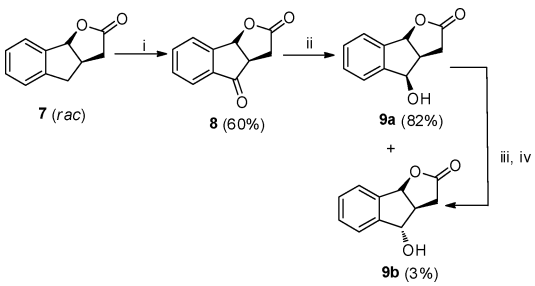
the bioactivity.[2,19,20] With the aim to a better understanding of the structure–activity relationship, the synthesis of the aromatic A-ring analogue of orobanchol, *viz.* GR24 having a hydroxy group in the B-ring, was undertaken. The synthetic route should thereby be chosen in such a manner that both C-4-OH epimers are accessible, as the spatial position of the OH group or its acetate may provide relevant information about the structure–activity requirements of the germination stimulatory activity of strigolactone analogues.

Interestingly, a strigolactone having an aromatic A-ring has recently been isolated from root exudates of tobacco.[19] The structure of this solanacol **6** was first incorrectly assigned. The methyl groups in the A-ring are postioned *ortho* instead of *para*.[21] The occurrence of a natural aromatic strigolactone is an extra stimulus for the present study.

Results and discussion

Synthesis

For the synthetic strategy of an aromatic A-ring analogue of orabanchol, the ABC skeleton of GR24 is a logical starting material. This compound (*viz. rac.* **7**) is readily available from either indan-1-one[22] or benzaldehyde.[23] Selective oxidation at the C-4 position could only be achieved after considerable experimentation (Scheme 1). It was found that the use of $FeCl_3$ with *tert*-butylhydroperoxide (TBHP) in pyridine[24] is most adequate resulting in 60% yield and 90% based on recovered starting material. Subsequent reduction of the thus obtained 4-oxo compound **8** with sodium borohydride under Luche conditions delivered a mixture of the *cis* and *trans* C-4-OH products, **9a** and **9b**, respectively, in a ratio of 82 : 3. The structure of the *cis* product **9a** was ascertained by X-ray diffraction analysis (Fig. 2). This analysis showed that the OH group was oriented *cis* to the C-ring, thereby confirming that hydride attack takes place primarily from the least hindered convex side.



Scheme 1 *Reagents and conditions*: (i) $FeCl_3$ (2 mol%), aq TBHP (70%, 3 equiv) pyridine, 82 °C, 24 h, (ii) $NaBH_4$, $CeCl_3{\cdot}7H_2O$, EtOH, (iii) PPh_3, $PhCO_2H$, $EtO_2CN{=}NCO_2Et$, toluene, (iv) K_2CO_3, MeOH.

In orobanchol, however, the OH has a *trans* orientation with respect to the lactone unit. In order to obtain the desired *trans*

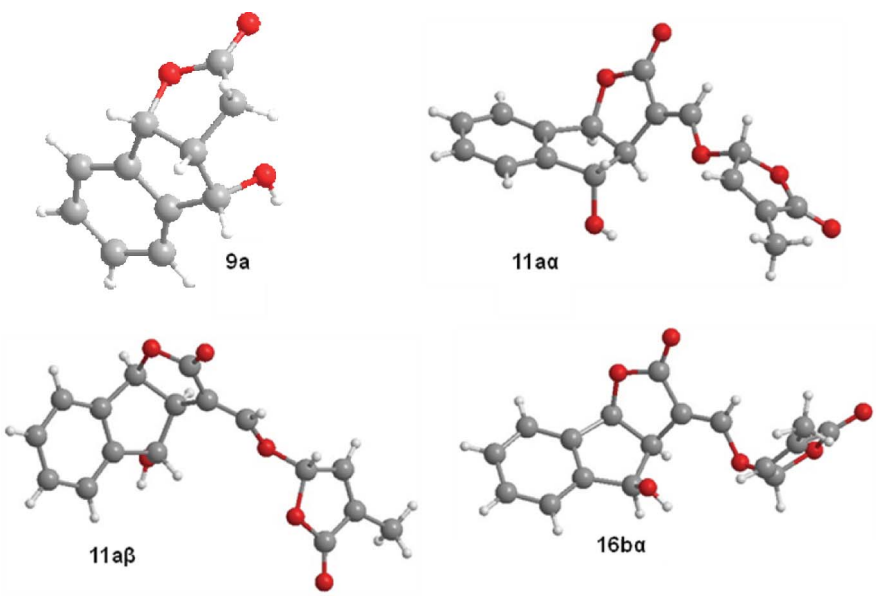
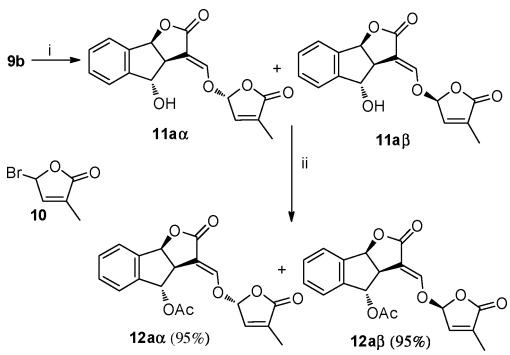


Fig. 2 Crystal structures of compounds **9a**, **11aα-β** and **16bα**.

product **9b** in better quantities, the *cis* compound **9a** was subjected to a Mitsunobu reaction, which afforded the *trans* product in 96% yield. Having both C-4-OH epimers **9a** and **9b** in hand, the next step was coupling with the D-ring.

To this end, the *trans* C-4-OH compound was formylated with ethyl formate in the presence of potassium *tert*-butoxide, followed by reaction with bromobutenolide **10** (Scheme 2).[22,23] This two-step one-pot process resulted in a mixture of two diastereomeric aromatic A-ring analogues of orobanchol, **11aα** and **11aβ** (ratio *ca* 1 : 1), in 78% overall yield. These diasteroisomers were readily separated by column chromatography, after which both structures were confirmed by X-ray diffraction analysis (Fig. 2).



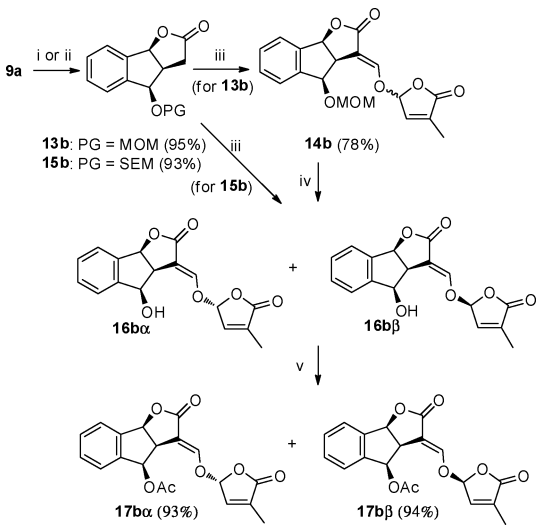
Scheme 2 *Reagents and conditions*: (i) HCO_2Me, *t*-BuOK, THF, then butenolide **10**, DME (ii) Ac_2O, pyridine, CH_2Cl_2.

Much to our surprise, direct coupling of *cis* alcohol **9a** with the D-ring using the one-pot two-step sequence did not meet with success, probably due to the fact that the spatial position of the free C-4 hydroxyl is in close proximity of the enolate anion. Initially, we tried to solve this problem by using the methoxymethyl (MOM) group as a protecting group, resulting in a smooth coupling with butenolide **10** in 78% yield (Scheme 3). However, deprotection required prolonged treatment with $ZnBr_2$ leading to a relatively low yield (*ca.* 60%) of the desired alcohols **16bα** and **16bβ** due to unreacted starting material and to decomposition. Next, we turned to the 2-trimethylsilylethoxymethyl (SEM) protecting group giving **15b** in 93% yield.

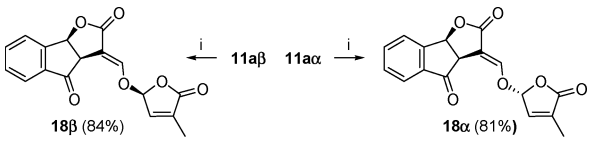
The subsequent coupling proceeded smoothly, after which deprotection was conveniently achieved by direct treatment of the reaction mixture with 1 M HCl. Via the latter pathway the *cis* aromatic A-ring analogues **16bα** and **16bβ** were obtained in a *ca.* 1 : 1 ratio in 74% yield starting from **15b** and 68% overall yield starting from **9a**. The structure of alcohol **16bα** was also confirmed by X-ray crystallographic analysis (Fig. 2).

The corresponding acetates **12aα** and **β** and **17bα** and **β** of all four diasteomeric aromatic A-ring analogues were prepared by straightforward acetylation of the hydroxyl groups in very good yields (Schemes 2 and 3). These aromatic A-ring orobanchyl acetates are in fact aromatic analogues of alectrol. The structure of this strigolactone was under discussion for quite a while,[16,25–27] but recently shown to be orobanchyl acetate.[20]

Furthermore, the *trans* alcohols **11aα** and **β** were also subjected to oxidation conditions using PDC in CH_2Cl_2 providing the corresponding ketones **18α** and **β** in 81% and 84% yield, respectively (Scheme 4). These analogues are also of biological interest as little is know about the activity of oxo containing strigolactones.



Scheme 3 *Reagents and conditions*: (i) MOM chloride, DIPEA, CH_2Cl_2, rt, 16 h, (ii) DIPEA, SEMCl, CH_2Cl_2, 22 h, (iii) HCO_2Me, *t*-BuOK, THF, then butenolide **10**, DME, workup with 1 M HCl (iv) $ZnBr_2$, CH_2Cl_2, 25 °C, (v) Ac_2O, pyridine, CH_2Cl_2.



Scheme 4 *Reagents and conditions*: (i) PDC, CH_2Cl_2.

The results reveal that that the oxidation route to introduce a hydroxyl group at the C-4 position of the B-ring in GR24 is quite effective. Takikawa *et al.*[21] used an entirely different approach to install a hydroxyl group in the B-ring in their synthesis of solanacol with the aim to prove its structure. These authors used a Diels–Alder reaction of a furanone as the first step following a literature procedure.[28] Our route is more direct.

Bioactivity

The newly prepared aromatic A-ring analogues of orobanchol were bioassayed against seeds of *Striga hermonthica* and *Orobanchae ramosa* using a standard protocol (see Fig. 3–6).[29] In all cases GR24 was used as the standard and water as the blank control. The bioassays for **11aα**, **11aβ**, **16bα** and **16bβ** on seeds of *O. ramosa* clearly reveal that **11aα** is the most active one. In this analogue the relative position of the C-4-OH and the D-ring with respect to the C-ring lactone is the same as in natural (+)-orobanchol (**3**). The same holds for the bioassays of this series of analogues for seeds of *S. hermonthica*. When the C-4-OH is positioned *cis* with respect to the C-ring lactone, the activities are markedly lower. This difference may be due to a favorable effect of the *trans*-C-4-OH on binding the stimulants in the receptor. The results suggest that *S. hermonthica* is more sensitive to structural changes than

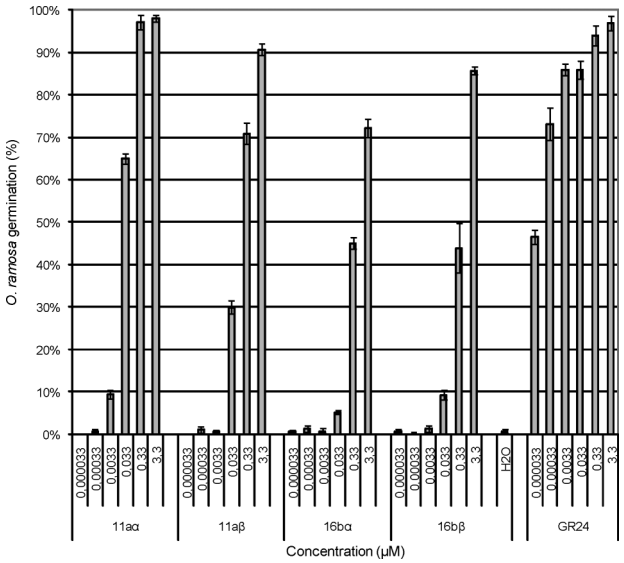


Fig. 3 Bioassay: germination activity towards seeds of *O. ramosa*.

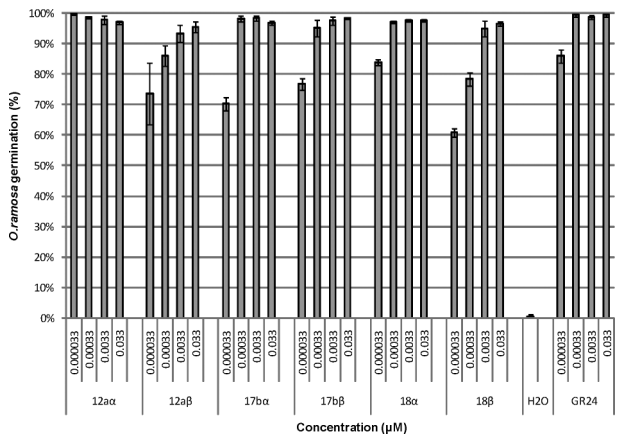


Fig. 4 Bioassay: germination activity towards seeds of *O. ramosa*.

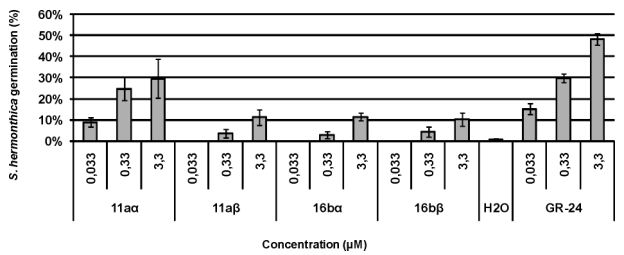


Fig. 5 Bioassay: germination activity towards seeds of *S. hermonthica*.

O. Ramosa. In the literature only a few examples are known in which all possible diastereoisomers of a particular strigolactone have been tested for germination of parasitic weed seeds.[14a,30–32] In case of strigolactone **2**, it was shown that the natural stereochemistry of the stimulant corresponds with the highest activity.[14a] The same was observed for strigol (**1**),[31] GR24[32] and demethylsorgolactone[30] (compound **2**, lacking the methyl group in the A-ring).

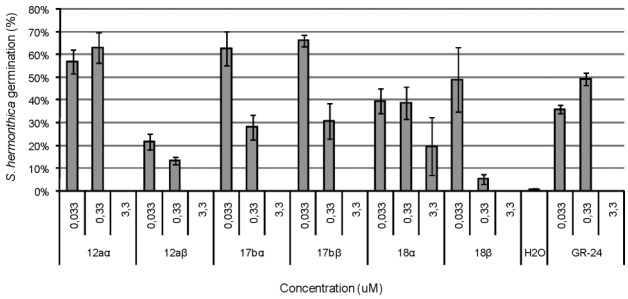


Fig. 6 Bioassay: germination activity towards seeds of *S. hermonthica*.

The bioactivity of all acetates is very high for *O. ramosa.* It seems that the spatial arrangement of the acetate group hardly influences the germination seeds of *O. ramosa.* This finding is remarkable as in natural strigolactones C-4-*O*-acetates score lower than the corresponding hydroxy compounds.[2,20] In the case of *S. hermonthica* seeds the C-4-*O*-acetate **12aα**, which has the same relative *syn* stereochemistry of the C-4-*O*-acetate and the D-ring as in natural orobanchyl acetate (alectrol), is considerably more active than **12aβ** having the *anti* stereochemistry. The activities of the stereoisomers **17bα** and **17bβ** which have the C-4-*O*-acetate in the unnatural position are remarkably high. Here the O-actylation enhances the activity (compare the activities of hydroxy compounds **16bα** and **16bβ** with those of the acetates **17bα** and **17bβ**).

The two oxo compounds **18α** and **18β** both are remarkably active in inducing germination of both seed types, but the isomers with the D-ring in the natural configuration perform best. These observations for the C-4-*O*-acetates and the C-4-oxo compounds may be an indication that hydrogen bonding of the C-4-OH is not an important aspect for the binding of the stimulant molecules in the receptor.

In summary, it may be concluded that the diasteromers of the newly prepared aromatic A-ring analogues of orobanchol possessing the natural relative configuration both at C-4 and at the D-ring are the most active ones. *S. hermonthica* seeds are more sensitive to structural changes than seeds of *O. ramosa*.

Experimental section

General remarks

All glass apparatus were oven dried prior to use. Solvents were distilled from appropriate drying agents prior to use and stored under nitrogen. Standard syringe techniques were applied for the transfer of dry solvents and air or moisture-sensitive reagents. All chemicals were obtained from commercial sources and used without further purification. IR spectra were recorded on an ATI Mattson Genesis Series FTIR spectrometer, or on a Bruker Tensor 27 FTIR spectrometer. Melting points were analyzed with a Buchi melting point apparatus B-545. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker DRX-300 (operating at 300 MHz for ^{1}H and at 75 MHz for ^{13}C spectra) spectrometer using deuterated solvents. Tetramethylsilane (0.00 ppm) served as an internal standard in ^{1}H NMR and $CDCl_3$ (77.0 ppm) in ^{13}C NMR spectra. Coupling constants are reported as *J*-values in Hz. Multiplicities are reported as: s (singlet), d (doublet), t (triplet),

dd (doublet of doublets), m (multiplet) in ^{1}H NMR spectra. Reactions were monitored using thin layer chromatography (TLC) on silica gel-coated plates (Merck 60 F254) with the indicated solvent mixture. Detection was performed with UV-light, and/or by charring at ~150 °C after dipping in to a solution of either 2% anisaldehyde in ethanol/H_2SO_4 or $(NH_4)_6Mo_7O_{24}\cdot 4H_2O$ (25 g L^{-1}) or $KMnO_4$. High resolution mass spectra were recorded on a JEOL AccuTOF (ESI), or a MAT900 (EI, CI, and ESI). Column chromatography was performed over silica gel (0.035–0.070 mm) using freshly distilled solvents. Air and moisture sensitive reactions were carried out under an inert atmosphere of dry nitrogen or argon.

(3a*R,8b*R**)-3,3a-Dihydro-2*H*-indeno[1,2-*b*]furan-2,4(8b*H*)-dione (8).** Compound **7** (5.01 g, 28.7 mmol) was added to the solution of $FeCl_3\cdot 6H_2O$ (0.15 g, 0.57 mol) in pyridine (30 mL). After the addition of *tert*-butyl hydroperoxide (70% in H_2O; 11.9 mL, 86.1 mmol), the reaction mixture was heated at 82 °C for 24 h. The mixture was then allowed to cool to room temperature and poured into 1 N aqueous HCl (200 mL) in order to remove the pyridine. The organic phase was extracted with Et_2O (2 × 200 mL), washed with brine and dried ($MgSO_4$). After filtration, the filtrate was concentrated under vacuum. The remaining mixture was separated by column chromatography (EtOAc/n-heptane 1 : 3) affording product **8** (3.24 g, 60% (90% based on recovered **7**)). Mp 113.5–114 °C. FT-IR (solid) cm^{-1}: 1757, 1713. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.84–7.82 (m, 1H), 7.78–7.76 (m, 2H), 7.63–7.58 (m, 1H), 6.00 (d, 1H, J = 6.6 Hz), 3.61–3.53 (m, 1H), 3.12–3.02 (m, 1H), 2.78 (dd, 1H, J = 19.2, 4.5 Hz).^{13}C NMR (75 MHz, $CDCl_3$): δ 202.0 (s), 174.3 (s), 149.2 (2 × s), 135.6 (d), 130.6 (d), 127.1 (d), 123.8 (d), 78.6 (d), 45.3 (d), 30.6 (t). HRMS (ESI) m/z calcd for $C_{11}H_8O_3$ (M+Na)$^+$: 211.03711, found: 211.03557.

(3a*S,4*R**,8b*R**)-4-Hydroxy-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-2-one (9a).** To a stirred solution of ketone **8** (3.00 g, 15.9 mmol) in ethanol (50 mL) was added $CeCl_3\cdot 7\ H_2O$ (5.94 g, 15.9 mmol) followed by slow addition of $NaBH_4$ (0.60 g, 15.9 mmol). After stirring for 10 min, the reaction mixture was quenched by dropwise addition of 1 M HCl and then extracted with dichloromethane (2 × 100 mL). The organic layer was washed with brine and dried over $MgSO_4$. Evaporation of the solvent gave a crude product, which was chromatographed on silica gel (EtOAc/n-heptane 1 : 1) to afford **9a** (2.48 g, 82%) as a white solid and **9b** (90 mg, 3%) as a colorless oil. The crystalline **9a** was crystallized from CH_2Cl_2–n-hexane (1 : 2) to give colorless crystals. Mp 154.3–154.8 °C. FT-IR (solid) cm^{-1}: 3399, 1766. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.49–7.34 (m, 4H), 5.73 (d, 1H, J = 7.0 Hz), 5.19 (d, 1H, J = 7.2 Hz), 3.53–3.43 (m, 1H), 2.79 (dd, 1H, J = 18.6, 6.0 Hz), 2.63 (dd, 1H, J = 18.6, 10.2 Hz).^{13}C NMR (75 MHz, $CDCl_3$): δ 178.1 (s), 144.5 (s), 137.8 (s), 129.4 (d), 128.3 (d), 125.1 (d), 124.8 (d), 84.6 (d), 72.0 (d), 42.6 (d), 27.9 (t). HRMS (ESI) m/z calcd for $C_{11}H_{10}O_3$ (M+Na)$^+$: 213.05276, found: 213.05124.

(3a*S,4*S**,8b*R**)-4-Hydroxy-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-2-one (9b).** To a stirred mixture of alcohol **9a** (1.00 g, 5.26 mmol), triphenylphosphine (4.82 g, 18.4 mmol) and benzoic acid (2.24 g, 18.4 mmol) in toluene (30 mL) was added a solution of diethyl azodicarboxylate (3.20 g, 18.4 mmol) in toluene (10 mL). The mixture was stirred for 14 h and then concentrated under reduced pressure. Silica gel chromatography (EtOAc–n-hexane 1 : 1) of the residue gave (1.46 g, 95%) corresponding benzoate as a colorless oil. A mixture of the benzoate (1.40 g, 4.75 mmol) and K_2CO_3 (0.99 g, 7.18 mmol) in MeOH (20 mL) was stirred for 1.5 h, acidified with 1 M HCl and then extracted with ethyl acetate (2 × 50 mL). The organic layer was washed with water and brine, and then dried over $MgSO_4$. Evaporation of the solvent gave a crude product, which was purified by silica gel column chromatography (EtOAc/n-heptane 1 : 1) to afford **9b** (1.31 g, 95%) as a colorless oil. FT-IR (thin film) cm^{-1}: 3398, 1767. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.43–7.34 (m, 4H), 5.91 (d, 1H, J = 6.0 Hz), 3.39 (bs, 1H), 3.13–3.05 (m, 1H), 2.80 (dd, 1H, J = 18.3, 10.5 Hz), 2.34 (dd, 1H, J = 18.3, 5.4 Hz). ^{13}C NMR (75 MHz, $CDCl_3$): δ 176.5 (s), 143.7 (s), 138.1 (s), 130.1 (d), 129.3 (d), 125.9 (d), 125.0 (d), 85.7 (d), 79.6 (d), 47.3 (d), 32.9 (t). HRMS (ESI) m/z calcd for $C_{11}H_{10}O_3$ (M+Na)$^+$: 213.05276, found: 213.05220.

5-Bromo-3-methylfuran-2(5*H*)-one (10). To 3-methylfuran-2(5*H*)-one (0.20 g, 2.06 mmol) in dry CCl_4 (20 mL) was added NBS (0.40 g, 2.26 mmol) and AIBN (5 mg) and the resulting reaction mixture was heated at reflux for 2 h while irradiating with a 250 W lamp. The mixture was cooled to 0 °C and solid succinimide was filtered off. The solvent was removed *in vacuo* to give bromobutenolide **10**, which was used as such in the coupling step.

(3a*S,4*R**,8b*S**,*E*)-4-Hydroxy-3-((((*R**)-4-methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-2-one (11aα) and its 2′*S** diastereomer (11aβ).** Potassium *tert*-butoxide (0.21 g, 1.89 mmol) was added in small portions to a solution of lactone **9b** (0.30 g, 1.57 mmol) and methyl formate (0.2 mL, 3.26 mmol) in anhydrous THF (10 mL, freshly distilled) while stirring at 0 °C under nitrogen. Stirring was continued at room temperature until all lactone had reacted (monitored by TLC, EtOAc/n-heptane 3 : 7). THF was removed *in vacuo*, after which the resulting salt was used as such in the coupling with bromobutenolide **10**. To the formylated product in DME (10 mL) was added a solution of bromobutenolide **10** in DME (5 mL). The reaction mixture was stirred overnight, DME was evaporated, then diluted with water (15 mL) and extracted with EtOAc (3 × 20 mL). The organic layer was dried (Na_2SO_4) and concentrated. The resulting diastereomeric mixture was purified by silica gel flash chromatography (EtOAc/n-heptane 1 : 1) to afford two partly separated diastereomeric products (**11aα** and **11bβ**, ratio *ca* 1 : 1, 0.38 g, 78%). The faster moving diastereomer was crystallized from CH_2Cl_2–n-hexane (1 : 3) to give **11aα** as colorless crystals. The slower moving diastereomer was also crystallized from CH_2Cl_2–n-hexane (1 : 3) to give **11aβ** as colorless crystals.

Diastereomers **16bα**, **16bβ** and **14b** were prepared by the same procedure. Products **16bα** and **16bβ** were obtained from **15b** in a *ca* 1 : 1 ratio in 74% yield.

11aα. Mp 179.5–180 °C. FT-IR (solid) cm^{-1}: 3452, 1779, 1744, 1674. ^{1}H NMR (300 MHz, CD_3CN): δ 7.58 (d, 1H, J = 2.7 Hz), 7.53–7.50 (m, 1H), 7.46–7.40 (m, 3H), 7.15–7.13 (m, 1H), 6.36–6.34 (m, 1H), 6.05 (d, 1H, J = 7.5 Hz), 5.23 (s, 1H), 3.73–3.69 (m,

1H), 1.96 (t, 3H, J = 1.5 Hz).^{13}C NMR (75 MHz, CD_3CN): δ 170.3 (s), 170.1 (s), 152.0 (d), 144.6 (s), 141.6 (d), 139.1 (s), 134.4 (s), 129.0 (d), 129.8 (d), 125.6 (d), 125.4 (d), 109.1 (s), 100.8 (d), 83.6 (d), 78.1 (d), 49.7 (d), 9.3 (q). HRMS (ESI) m/z calcd for $C_{17}H_{14}O_6$ $(M+Na)^+$: 337.06881, found: 337.06913. **11aβ**: Mp 73.7–78.2 °C. FT-IR (solid) cm^{-1}: 3446, 1782, 1739, 1670. ^{1}H NMR (300 MHz, CD_3CN): δ 7.57–7.56 (d, 1H), 7.52–7.49 (m, 1H), 7.44–7.41 (m, 3H), 7.13 (t, 1H, J = 3.0 Hz), 6.36 (t, 1H, J = 2.4 Hz), 6.06 (d, 1H, J = 7.2 Hz), 5.17 (bs, 1H), 3.74–3.70 (m, 1H), 1.96 (t, 3H, J = 2.7 Hz).^{13}C NMR (75 MHz, CD_3CN): δ 170.3 (s), 170.1 (s), 151.9 (d), 144.6 (s), 141.5 (d), 139.0 (s), 134.5 (s), 129.8 (d), 128.9 (d), 125.5 (d), 125.4 (d), 109.3 (s), 100.8 (d), 83.6 (d), 78.2 (d), 49.6 (d), 9.2 (q). HRMS (ESI) m/z calcd for $C_{17}H_{14}O_6$ $(M+Na)^+$: 337.06881, found: 337.06907.

(3a*S,4*R**,8b*S**,*E*) - 4 - (Methoxymethoxy) - 3 - ((((*R**,*S**)-4-methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-3,3a,4,8b-tetrahydro-2*H*-indeno[1,2-*b*]furan-2-one (14b).** Yield: 78% (colorless oil). FT-IR (film) cm^{-1}: 2954, 1782, 1744, 1683.^{1}H NMR (300 MHz, $CDCl_3$): δ 7.60 (dd, 1H, J = 7.8, 2.1 Hz), 7.55–7.53 (m, 1H), 7.46–7.37 (m, 3H), 6.93 (q, 1H, J = 1.5 Hz), 6.20–6.17 (m, 1H), 5.61 (dd, 1H, J = 7.2, 2.1 Hz), 5.24–5.18 (m, 1H), 4.79–4.53 (m, 2H), 4.13–3.99 (m, 1H), 3.42 & 3.38 (s, 3H), 2.01–2.00 (m, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.6 and 170.5 (s), 169.7 and 169.6 (s), 150.8 and 150.7 (d), 142.8 and 142.6 (s), 140.5 and 140.3 (d), 137.9 (s), 135.6 and 135.3 (s), 130.1 (d), 129.1 (d), 126.0 (d), 108.5 and 108.4 (s), 100.3 and 100.2 (d), 96.2 (2 × t), 87.7 (2 × d), 79.4 and 79.2 (d), 55.3 (2 × q), 43.9 (d), 10.2 (q). HRMS (ESI) m/z calcd for $C_{19}H_{18}O_7$ $(M+Na)^+$: 381.09502, found: 381.09357.

(3a*S,4*S**,8b*S**,*E*)-4-Hydroxy-3-((((*R**)-4-methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methyl ene)-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-2-one (16bα).** Mp 175.8–176.3 °C. FT-IR (solid) cm^{-1}: 3394, 1787, 1731, 1666. ^{1}H NMR (300 MHz, CD_3CN): δ 7.63 (d, 1H, J = 2.1 Hz), 7.55–7.52 (m, 1H), 7.48–7.38 (m, 3H), 7.11–7.09 (m, 1H), 6.35–6.34 (m, 1H), 5.68 (d, 1H, J = 3.7 Hz), 5.27 (t, 1H, J = 14.1 Hz), 4.06 (td, 1H, J = 7.8, 2.4 Hz), 3.40 (d, OH, J = 6.9 Hz), 1.95 (t, 3H, J = 1.5 Hz). ^{13}C NMR (75 MHz, CD_3CN): δ 170.6 (s), 170.3 (s), 151.6 (d), 145.1 (s), 141.6 (d), 138.1 (s), 134.4 (s), 129.8 (d), 128.7 (d), 125.6 (d), 125.5 (d), 107.6 (s), 100.6 (d), 82.8 (d), 73.3 (d), 44.7 (d), 9.2 (q). HRMS (ESI) m/z calcd for $C_{17}H_{14}O_6$ $(M)^+$: 315.08686, found: 315.08754.

(3a*S,4*S**,8b*S**,*E*)-4-Hydroxy-3-((((*S**)-4-methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-2-one (16bβ).** Mp 218.8–219.3 °C. FT-IR (solid) cm^{-1}: 3334, 1786, 1730, 1668. ^{1}H NMR (300 MHz, DMSO-d_6): δ 7.75 (d, 1H, J = 2.1 Hz), 7.49 (d, 1H, J = 6.9 Hz), 7.42–7.34 (m, 3H), 6.66 (bs, 1H), 5.65 (d, 1H, J = 6.9 Hz), 5.37 (d, 1H, J = 6.9 Hz), 5.16 (t, 1H, J = 7.5 Hz), 3.96 (td, 1H, J = 7.8, 2.1 Hz), 1.89 (s, 3H). ^{13}C NMR (75 MHz, DMSO-d_6): δ 171.0 (s), 170.8 (s), 152.9 (d), 146.4 (s), 143.4 (d), 138.3 (s), 133.4 (s), 130.0 (d), 128.7 (d), 126.0 (2 × d), 107.8 (s), 101.2 (d), 82.9 (d), 73.1 (d), 44.7 (d), 10.1 (q). HRMS (ESI) m/z calcd for $C_{17}H_{14}O_6$ $(M+Na)^+$: 337.06881, found: 337.06939.

(3a*S,4*S**,8b*S**,*E*)-3-((((*R**)-4-Methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-2-oxo-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-4-yl acetate (12aα).** To a solution of **11aα** (0.10 g, 0.31 mmol) in dichlomethane (5 mL) was added pyridine (1 mL), DMAP (catalytic) and acetic anhydride (0.5 mL) at 0° C. The reaction mixture was allowed to warm to room temperature and stirred for 2 h. Solvent was removed *in vacuo* and the residue was purified by by silica gel flash chromatography (EtOAc/n-heptane 1 : 2) to afford **12aα** (0.10 g, 95%) as a white solid. Mp 190.8–191.3 °C. FT-IR (solid) cm^{-1}: 3457, 1781, 1738, 1676. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.59 (d, 1H, J = 2.7 Hz), 7.55–7.53 (m, 1H), 7.47–7.40 (m, 3H), 7.01–7.00 (m, 1H),, 6.44 (bs, 1H), 6.18–6.17 (m, 1H), 6.10 (d, 1H, J = 7.5 Hz), 3.84–3.88 (m, 1H), 2.05 (s, 3H), 2.02 (t, 3H, J = 2.7 Hz). ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.8 (s), 169.7 (s), 169.6 (s), 152.5 (d), 140.6 (d), 140.1 (s), 140.0 (s), 135.1 (s), 130.1 (d), 130.0 (d), 126.0 (d), 125.9 (d), 108.2 (s), 100.1 (d), 83.2 (d), 78.4 (d), 46.7 (d), 20.6 (q), 10.2 (q). HRMS (ESI) m/z calcd for $C_{19}H_{16}O_7$ $(M+Na)^+$: 379.07937, found: 379.07731.

Compounds **12aβ**, **17bα** and **17bβ** were prepared following the same procedure.

(3a*S,4*R**,8b*S**,*E*)-3-((((*R**)-4-Methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-2-oxo-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-4-yl acetate (12aβ).** Yield: 95%. Mp 167.3–167.8 °C. FT-IR (solid) cm^{-1}: 3442, 1769, 1729, 1660.^{1}H NMR (300 MHz, $CDCl_3$): δ 7.54–7.50 (m, 2H), 7.46–7.39 (m, 3H), 6.98–6.96 (m, 1H), 6.37 (bs, 1H), 6.21–6.20 (m, 1H), 6.10 (d, 1H, J = 7.2 Hz), 3.85–3.89 (m, 1H), 2.03–2.02 (m, 6H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 169.7 (s), 169.6 (s), 169.4 (s), 151.3 (d), 140.3 (d), 140.0 (2 × s), 135.7 (s), 130.1 (d), 130.0 (d), 126.2 (d), 125.8 (d), 108.5 (s), 99.5 (d), 83.1 (d), 78.5 (d), 46.8 (d), 20.6 (q), 10.3 (q). HRMS (ESI) m/z calcd for $C_{19}H_{16}O_7$ $(M+Na)^+$: 379.07937, found: 379.07745.

(3a*S,4*S**,8b*S**,*E*)-3-((((*R**)-4-Methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-2-oxo-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-4-yl acetate (17bα).** Yield: 93%. Mp 160.4–160.9 °C. FT-IR (solid) cm^{-1}: 2958, 1782, 1731, 1687. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.58–7.56 (m, 2H), 7.46–7.37 (m, 3H), 6.95–6.93 (m, 1H), 6.54 (d, 1H, J = 8.1 Hz), 6.16–6.14 (m, 1H), 5.70 (d, 1H, J = 7.2 Hz), 4.19–4.13 (m, 1H), 2.03 (s, 3H), 2.01 (t, 3H, J = 1.5 Hz). ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.1 (s), 170.0 (s), 169.5 (s), 150.7 (d), 140.8 (d), 140.4 (s), 138.5 (s), 135.7 (s), 130.4 (d), 129.7 (d), 126.1 (d), 125.7 (d), 107.8 (s), 99.6 (d), 82.6 (d), 73.6 (d), 43.2 (d), 20.7 (q), 10.3 (q). HRMS (ESI) m/z calcd for $C_{19}H_{16}O_7$ $(M+Na)^+$: 379.07937, found: 379.07807.

(3a*S,4*R**,8b*S**,*E*)-3-((((*S**)-4-Methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-2-oxo-3,3a,4,8b-tetrahydro-2*H*-indeno-[1,2-*b*]furan-4-yl acetate (17bβ).** Yield: 94%. Mp 158.1–158.6 °C. FT-IR (solid) cm^{-1}: 2958, 1774, 1735, 1683. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.64 (d, 1H, J = 2.4 Hz), 7.59–7.56 (m, 1H), 7.47–7.37 (m, 3H), 6.94–6.92 (m, 1H), 6.53 (d, 1H, J = 8.4 Hz), 6.15–6.14 (m, 1H), 5.72 (d, 1H, J = 7.5 Hz), 4.20–4.14 (m, 1H), 2.03 (t, 3H, J = 1.5 Hz), 2.00 (bs, 3H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 170.1 (s), 169.7 (s), 169.5 (s), 151.8 (d), 140.9 (s), 140.1 (d), 138.4 (s), 135.3 (s), 130.4 (d), 129.7 (d), 126.0 (d), 125.7 (d), 107.2 (s), 100.0 (d), 82.5 (d), 73.2 (d), 42.9 (d), 20.4 (q), 10.3 (q). HRMS (ESI) m/z calcd for $C_{19}H_{16}O_7$ $(M+Na)^+$: 379.07937, found: 379.07693.

(3a*S,4*R**,8b*S**)-4-(Methoxymethoxy)-3,3a,4,8b-tetrahydro-2*H*-indeno[1,2-*b*]furan-2-one (13b).** To a solution of compound **9a** (0.51 g, 2.68 mmol) in dichloromethane (15 mL) was added *N*,*N*-diisopropylethylamine (0.7 mL, 4.01 mmol) and MOMCl (0.3 mL, 3.85 mmol) at 25 °C for 3 h. Solvent was removed *in vacuo* and the residue was purified by silica gel column chromatography (EtOAc/n-heptane 1 : 4) to afford **13b** (0.59 g, 95%) as a white

solid. Mp 84.3–84.8 °C. FT-IR (solid) cm^{-1}: 2954, 1757. ^1H NMR (300 MHz, $CDCl_3$): δ 7.49–7.35 (m, 4H), 5.68 (d, 1H, J = 7.2 Hz), 5.13 (d, 1H, J = 6.9 Hz), 4.82–4.75 (m, 2H), 3.64–3.54 (m, 1H), 3.44 (s, 3H), 2.76–2.52 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 176.3 (s), 141.5 (s), 137.5 (s), 129.6 (d), 129.0 (d), 125.6 (d), 125.1 (d), 96.1 (t), 83.2 (d), 78.1 (d), 55.6 (q), 42.4 (t), 28.7 (d). HRMS (ESI) m/z calcd for $C_{13}H_{14}O_4$ $(M+Na)^+$: 257.07898, found: 257.07712.

(3a*S,4*R**,8b*S**)-4-((2-(Trimethylsilyl)ethoxy)methoxy)-3,3a,4,8b-tetrahydro-2*H*-indeno[1,2-*b*]furan-2-one (15b).** To a solution of compound **9a** (0.51 g, 2.68 mmol) in anhydrous dichloromethane (15 mL) was added *N*,*N*-diisopropylethylamine (2.74 mL, 15.72 mmol) and [β-(trimethylsilyl)ethoxy]methyl chloride (1 mL, 5.65 mmol), and the mixture was stirred under a nitrogen atmosphere for 22 h. The solution was diluted with dichloromethane and washed successively with 0.5 N aqueous HCl (2 × 20 mL) and water (2 × 20 mL). The organic layer was dried (Na_2SO_4), evaporated, and the resulting yellow syrup was purified by silica gel column chromatography (EtOAc/n-heptane 1 : 4) to afford **15b** (0.79 g, 93%) as a white solid. Mp 57.8–58.3 °C. FT-IR (solid) cm^{-1}: 2954, 1765. ^1H NMR (300 MHz, $CDCl_3$): δ 7.48–7.42 (m, 4H), 5.71–5.69 (m, 1H), 5.18–5.16 (m, 1H), 4.88–4.80 (m, 2H), 3.75–3.58 (m, 3H), 2.76–2.50 (m, 2H), 1.01–0.89 (m, 2H), 0.03 (m, 9H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 176.3 (s), 141.7 (s), 137.4 (s), 129.6 (d), 128.9 (d), 125.6 (d), 125.0 (d), 94.2 (t), 83.2 (d), 77.8 (d), 65.4 (t), 42.4 (d), 28.7 (t), 17.7 (t), -1.8 (3 × q). HRMS (ESI) m/z calcd for $C_{17}H_{24}O_4Si$ $(M+Na)^+$: 343.13415, found: 343.13189.

(3a*R,8b*S**,*E*)-3-((((*R**)-4-Methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-3,3a-dihydro-2*H*-indeno[1,2-*b*]furan-2,4(8b*H*)-dione (18α).** To a solution of **11aα** (0.100 g, 3.18 mmol) in dichloromethane (10 mL) was added pyridinium dichromate (PDC, 0.179 g, 4.77 mmol). The reaction mixture was stirred at room for 5 h, diluted with ether and filtered through celite. The solvent was evaporated and the residue was chromatographed (EtOAc/n-heptane 1 : 4) to afford **18α** (80 mg, 81%) as a white solid. Mp 256.3–256.8 °C. FT-IR (solid) cm^{-1}: 1778, 1752, 1713, 1670. ^1H NMR (300 MHz, DMSO-d_6): δ 7.88 (d, 1H, J = 2.4 Hz), 7.87–7.81 (m, 2H), 7.73–7.70 (m, 1H), 7.67–7.62 (m, 1H), 7.40–7.39 (m, 1H), 6.73–6.72 (m, 1H), 6.05 (d, 1H, J = 6.6 Hz), 4.34 (dd, 1H, J = 6.6, 2.4 Hz), 1.91 (bs, 3H). ^{13}C NMR (75 MHz, DMSO-d_6): δ 198.5 (s), 170.7 (s), 169.5 (s), 155.2 (d), 149.6 (s), 143.1 (d), 136.0 (d), 135.6 (s), 133.7 (s), 131.0 (d), 127.6 (d), 123.8 (d), 104.4 (s), 101.4 (d), 77.0 (d), 48.7 (d), 10.1 (q). HRMS (ESI) m/z calcd for $C_{19}H_{18}O_5$ $(M+Na)^+$: 335.05316, found: 335.05051.

Compound **18β** was prepared by the same procedure starting from **11aβ**.

(3a*R,8b*S**,*E*)-3-((((*R**)-4-Methyl-5-oxo-2,5-dihydrofuran-2-yl)oxy)methylene)-3,3a-dihydro-2*H*-indeno[1,2-*b*]furan-2,4(8b*H*)-dione (18β).** Yield: 84%. Mp 80.1–80.6 °C. FT-IR (solid) cm^{-1}: 1774, 1757, 1718, 1670. ^1H NMR (300 MHz, $CDCl_3$): δ 7.81–7.72 (m, 3H), 7.61–7.56 (m, 2H), 7.03–7.02 (m, 1H), 6.45–6.44 (m, 1H), 5.96 (d, 1H, J = 6.6), 4.28 (dd, 1H, J = 6.6, 2.4 Hz), 2.03 (m, 3H, J = 1.5 Hz). ^{13}C NMR (75 MHz, $CDCl_3$): δ 197.6 (s), 169.6 (s), 169.2 (s), 152.1 (d), 148.8 (s), 140.8 (d), 135.6 (d), 135.4 (s), 133.3 (s), 130.6 (d), 127.0 (d), 124.1 (d), 104.7 (s), 99.0 (d), 76.4 (d), 48.6 (d), 10.3 (q). HRMS (ESI) m/z calcd for $C_{19}H_{18}O_5$ $(M+Na)^+$: 335.05316, found: 335.04990.

Bioassay

The germination bioassays were conducted as reported earlier.[22,26,29] *Striga hermonthica* seeds were conditioned and then incubated with stimulant solution. Three to six concentrations were used (see Fig. 3–6). The number of germinated seeds was counted under a microscope. All tests were carried out in triplicate. The bar diagrams in Fig. 3–6 show the average values with the standard deviation.

Notes and references

1 K. Yoneyama, X. Xie, K. Yoneyama and Y. Takeuchi, *Pest Manag. Sci.*, 2009, **65**, 467–470 and refs cited therein.
2 X. Xie, K. Yoneyama and K. Yoneyama, *Annu. Rev. Phytopathol.*, 2010, **48**, 93–117.
3 K. Yoneyama, A. A. Awad, X. Xie, K. Yoneyama and Y. Takeuchi, *Plant Cell Physiol.*, 2010, **51**, 1095–1103.
4 K. Akiyama, K.-I. Matsuzaki and H. Hayashi, *Nature*, 2005, **435**, 824–827.
5 H. J. Bouwmeester, C. Roux, J. A. Lopez-Raez and G. Bécard, *Trends Plant Sci.*, 2007, **12**, 224–230.
6 A. Besserer, V. Puech-Pagès, P. Kiefer, V. Gomez-Roldan, A. Jauneua, S. Roy, J-C. Portais, C. Roux, G. Bécard and N. Séjalon-Delmas, *PLoS Biology*, 2006, **41**, 1239–1247.
7 K. Akiyama, S. Ogasawara, S. Ito and H. Hayashi, *Plant Cell Physiol.*, 2010, **51**, 1104–1117.
8 V. Gomez-Roldan, S. Fermas, P. B. Brewer, V. Puech-Pages, E. A. Dun, J.-P. Pillot, F. Letisse, R. Matusova, S. Danoun, J.-C. Portais, H. Bouwmeester, G. Becard, C. A. Beveridge, C. Rameau and S. F. Rochange, *Nature*, 2008, **455**, 189–194.
9 M. Umehara, A. Hanada, S. Yoshida, K. Akiyama, T. Arite, N. Takeda-Kamiya, H. Magome, Y. Kamiya, K. Shirasu, K. Yoneyama, J. Kyozuka and S. Yamaguchi, *Nature*, 2008, **455**, 195–200.
10 Y. Tsuchiya and P. McCourt, *Curr. Opin. Plant Biol.*, 2009, **12**, 556–561.
11 A. J. Humphrey and M. H. Beale, *Phytochemistry*, 2006, **67**, 636–640.
12 C. Y. Chen, J. H. Zou, S. Y. Zhang, D. Zaitlin and L. H. Zhu, *Sci. China, Ser. C: Life Sci.*, 2009, **52**, 693–700.
13 (*a*) C. E. Cook, L. P. Whichard, B. Turner, M. E. Wall and G. H. Egley, *Science*, 1966, **154**, 1189–1190; (*b*) D. W. Brooks, H. S. Bevinakatti and D. R. Powell, *J. Org. Chem.*, 1985, **50**, 3779–3781.
14 (*a*) Y. Sugimoto, S. C. M. Wigchert, J. W. J. F. Thuring and B. Zwanenburg, *J. Org. Chem.*, 1998, **63**, 1259–1267; (*b*) J. Matsui, M. Bando, M. Kido, Y Takeuchi and K. Mori, *Eur. J. Org. Chem.*, 1999, 2193–2194.
15 A. Reizelman and B. Zwanenburg, *Eur. J. Org. Chem.*, 2002, 810–814.
16 B. Zwanenburg, A. S. Mwakaboko, A. Reizelman, G. Anilkumar and D. Sethumadhavan, *Pest Manag. Sci.*, 2009, **65**, 478–491.
17 A. W. Johnson, G. Gowda, A. Hassanali, J. Knox, S. Monaco, Z. Razawi and G. Roseberry, *J. Chem. Soc., Perkin Trans. 1*, 1981, 1734–1743.
18 E. M. Mangnus and B. Zwanenburg, *J. Agric. Food Chem.*, 1992, **40**, 1066–1070.
19 X. Xie, D. Kusomoto, Y. Takeuchi, K. Yoneyama, Y. Yamada and K. Yoneyama, *J. Agric. Food Chem.*, 2007, **55**, 8067–8072.
20 X. Xie, K. Yoneyama, D. Kusumoto, Y. Yamada, T. Yokota, Y. Takeuchi and K. Yoneyama, *Phytochemistry*, 2008, **69**, 427–431.
21 H. Takikawa, S. Jikumaru, Y. Sugimoto, X. Xie, K. Yoneyama and M. Sasaki, *Tetrahedron Lett.*, 2009, **50**, 4549–4551.
22 E. M. Mangnus, F. J. Dommerholt, R. L. P. de Jong and B. Zwanenburg, *J. Agric. Food Chem.*, 1992, **40**, 1230–1235.
23 H. Malik, F. P. J. T. Rutjes and B. Zwanenburg, *Tetrahedron*, 2010, **66**, 7198–7203.
24 M. Nakanishi and C. Bolm, *Adv. Synth. Catal.*, 2007, **349**, 861–864.
25 K. Mori, *Eur. J. Org. Chem.*, 1998, 1479–1489.
26 S. C. M. Wigchert, E. Kuiper, G. J. Boelhouwer, G. H. L. Nefkens, J. A. C. Verkleij and B. Zwanenburg, *J. Agric. Food Chem.*, 1999, **47**, 1705–1710.
27 (*a*) K. Mori, J. Matsui, M. Bando, M. Kido and Y. Takeuchi, *Tetrahedron Lett.*, 1998, **39**, 6023–6026; (*b*) J. Matsui, M. Bando, M. Kido, Y. Takeuchi and K. Mori, *Eur. J. Org. Chem.*, 1999, 2195–2199.
28 D. R. Buckle, N. J. Morgan, J. W. Ross, H. Smith and B. A. Spicer, *J. Med. Chem.*, 1973, **16**, 1334–1339.

29 E. M. Mangnus, P. L. A. Stommen and B. Zwanenburg, *J. Plant Growth Regul.*, 1992, **11**, 91–98.
30 J. W. J. F. Thuring, N. W. J. T. Heinsman, R. W. A. W. M. Jacobs, G. H. L. Nefkens and B. Zwanenburg, *J. Agric. Food Chem.*, 1997, **45**, 507–513.
31 (*a*) A. Reizelman, M. Scheeren, G. H. L. Nefkens and B. Zwanenburg, *Synthesis*, 2000, 1944–1951; (*b*) A. Reizelman, *PhD Thesis*, University of Nijmegen, 2003, Chapter 2.
32 J. W. J. F. Thuring, G. H. L. Nefkens and B. Zwanenburg, *J. Agric. Food Chem.*, 1997, **45**, 2278–2283.