Synthesis of All Eight Stereoisomers of the Germination Stimulant Sorgolactone

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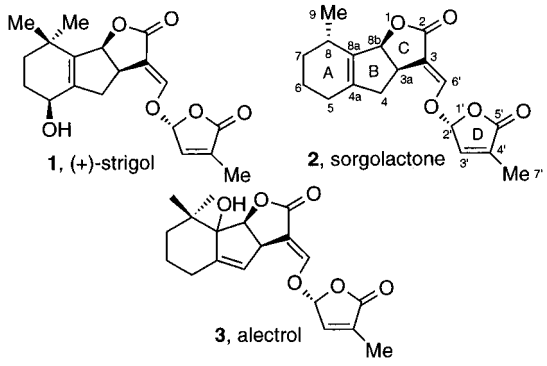
The naturally occurring sesquiterpene sorgolactone (**2**) belongs to the class of "strigolactones", which are highly potent germination stimulants for seeds of the parasitic weeds *Striga* and *Orobanche*. The aim of the present work was to synthesize all eight stereoisomers of sorgolactone and to evaluate their activities in the stimulation of germination of *S. hermontica* and *O. crenata*. Two racemic diastereomers of the ABC part of sorgolactone, **rac.10a** and **rac.10b** respectively, were prepared and coupled with homochiral latent D-ring synthons **12** and **ent.12**. In this manner, four mixtures of two separable (protected) sorgolactone diastereomers were obtained. Deprotection gave all eight target compounds as single isomers. Bioassays revealed that only those isomers possessing the same stereochemistry as natural sorgolactone at two adjacent chiral centers exhibit high biological activities.

Parasitic weeds of the genera *Striga*, *Orobanche*, and *Alectra* have an extremely devastating impact on several graminaceous and leguminous crops in tropical and semitropical areas of the eastern hemi-sphere.[1,2] These root parasitic weeds specifically interact with their hosts at four levels: (1) germination of the parasitic seed, (2) initiation of haustorial development, (3) transfer of water and minerals, and (4) host responses to infection.[3] The first two events mentioned require host-derived signals as recognition cues. Especially the stimulation of germination and the compounds that trigger this process have attracted much attention. The first naturally occurring germination stimulant, strigol (**1**) (Chart 1), was isolated from the root exudate of the false host cotton (*Gossypium hirsutum* L.)[4] and its structure was elucidated in 1972.[5] The absolute configuration was unambiguously assigned several years later.[6]

It was not until 1992 that some germination stimulants closely related to strigol were isolated from the root exudates of true hosts, viz. sorgolactone (**2**)[7] from sorghum (*Sorghum bicolor* (L.) Moench) and alectrol (**3**)[8] from cowpea (*Vigna unguiculata* (L.). Walp) (Chart 1). Strigol itself was shown to be the major *Striga* germination stimulant produced by maize (*Zea mays* L.) and proso millet (*Panicum miliaceum* L.)[9]

Chart 1



The structure of sorgolactone (**2**) has been tentatively assigned as depicted in Chart 1. On the basis of spectroscopic evidence its gross structure was elucidated. The absolute configuration of the stereogenic centers C3a, C8b, and C2′ was deduced from the similar circular dichroic (CD) data by comparison with those of (+)-strigol (**1**).

Because of lack of spectroscopic detail due to the small amount of sorgolactone obtained (5 μg from 300 000 sorghum plants) and the extremely laborious isolation procedure, the proposed structure could not be ascertained. Therefore the total synthesis of **2** was undertaken[10] to verify this tentative structure and to unambiguously establish the absolute stereochemical configuration. Very recently the synthesis of four racemic stereoisomers of sorgolactone was reported, supporting the gross structure proposed by Hauck.[11] Our goal was

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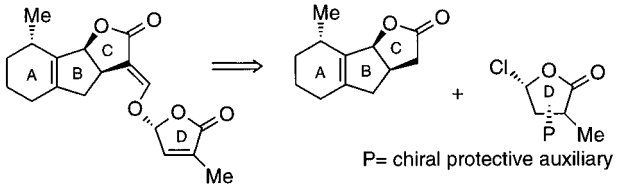
Scheme 1



to develop a strategy that allows the synthesis of all eight possible sorgolactone diastereoisomers as single isomers. Evaluation of their respective bioactivities will provide more insight in the structure−activity relationships of these germination stimulants. In this paper a full account of the synthesis and biological evaluation of these eight sorgolactones is presented.

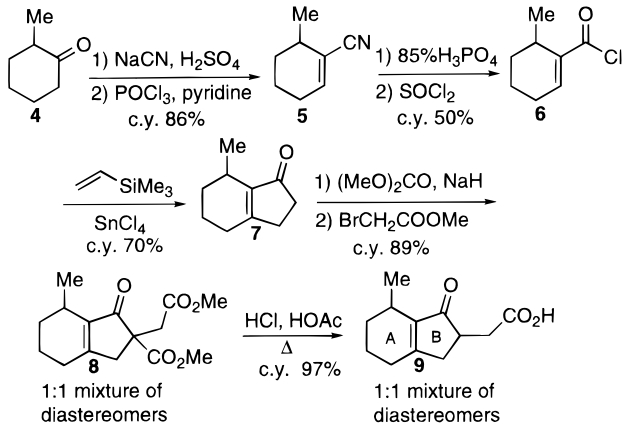
Results and Discussion

Retrosynthesis of the target compounds involves coupling of the D ring to the ABC part of the molecule (Scheme 1). Stereoselective coupling of the butenolide to diastereomerically pure tricyclic ABC lactone **10** furnishes a separable mixture of two protected sorgolactone diastereomers.

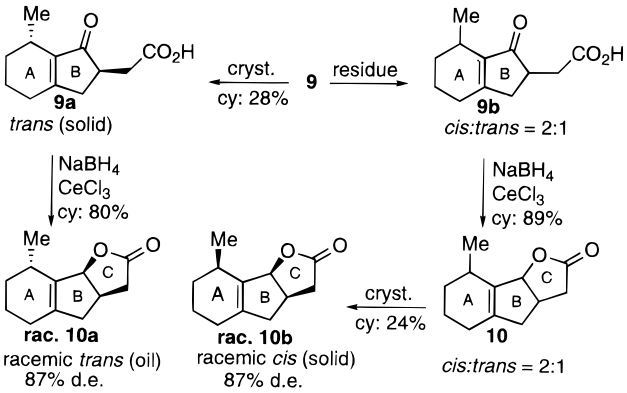
A method to introduce the D ring of a strigol analogue stereoselectively was recently developed in our laboratory[12,13] and applied in the stereoselective synthesis of strigol analogues GR7[12] and GR24[14] and demethylsorgolactone (DMSL), the sorgolactone analogue lacking the A-ring methyl group.[15] Application of this coupling methodology in the synthesis of **2** necessitates the preparation of trans and cis tricyclic lactones **rac.10a** and **rac.10b** as individual diastereomers.

To this end, acid chloride **6** was prepared via slightly modified literature procedures,[16−19] and was then subjected to a Friedel−Crafts reaction in the presence of trimethylvinylsilane, followed by a Nazarov cyclization of the in situ formed cyclopentadienyl cation to give AB fragment **7**[20] (Scheme 2). Thus obtained bicyclopentenone **7** was then converted via standard procedures[21] into keto acid **9** as a 1:1 mixture of diastereomers from which the trans isomer **9a** remained as a precipitate when **9** was dissolved in diisopropyl ether. Repeated crystallizations afforded the trans isomer of **9** in a diastereomeric excess of 87% as was deduced from a detailed analysis of the ^{13}C NMR spectra. The residue **9b**, enriched with the cis diastereomer, was subjected to a reduction by employing sodium borohydride in the presence of cerium trichloride,[22] which resulted in a mixture of the sorgolactone ABC fragments **10** (Scheme 3).

Scheme 2



Scheme 3



The cis tricyclic lactone **rac.10b** crystallized from the mixture and could be obtained in 87% de. Reduction of trans-AB-intermediate **9a** with sodium borohydride proceeded with complete retention of stereochemistry, leading to the trans-sorgolactone ABC part **rac.10a** in a purity of 93.5%. The contaminant was identified as the cis-ABC-fragment **10b**; consequently the de of **10a** is 87%. Using 2D-NOESY experiments, the relative configurations of **rac.10a** and **rac.10b** were assigned. The oily diastereomer **rac.10a** showed its characteristic doublet, which was attributed to H8, at 5.5 ppm, whereas the corresponding proton of crystalline **rac.10b** resonates at 5.3 ppm.

To obtain four racemic mixtures of the sorgolactone stereoisomers, both racemic ABC diastereomers **rac.10a** and **rac.10b** were coupled with chlorobutenolide **11**.[23,24] The reaction of **rac.10a** (of which only the 3a(*R*),8(*S*), 8b(*S*) enantiomer is drawn) with **11** gave a mixture of two diastereomeric racemates of sorgolactone, **rac.2a** and **rac.2b**, respectively (yields not optimized), in a ratio of about 1:1 (Scheme 4). They were readily separated by flash chromatography. In the same manner **rac.2c** and **rac.2d** were obtained starting from **rac.10b**, of which only the 3a(*R*),8(*R*),8b(*S*) enantiomer is depicted (Scheme 5).

The chemical shifts of the proton H8b of diastereoisomers **rac.2a**−**d** were compared with the corresponding value reported by Hauck.[7] In the trans-ABC sorgolactone diastereomers **rac.2a** and **rac.2b** the H8b doublet reso-

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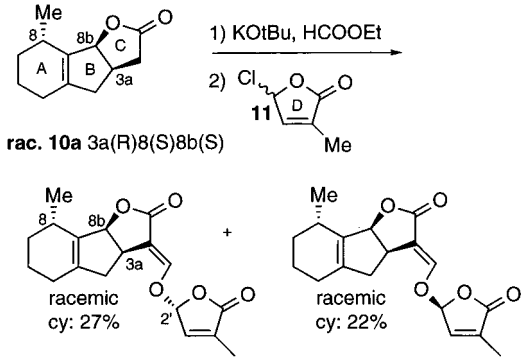
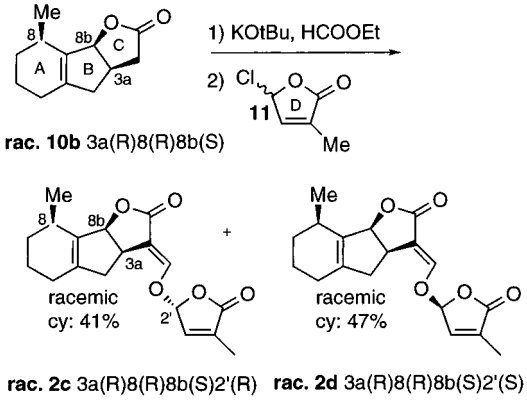
Scheme 4



Scheme 5

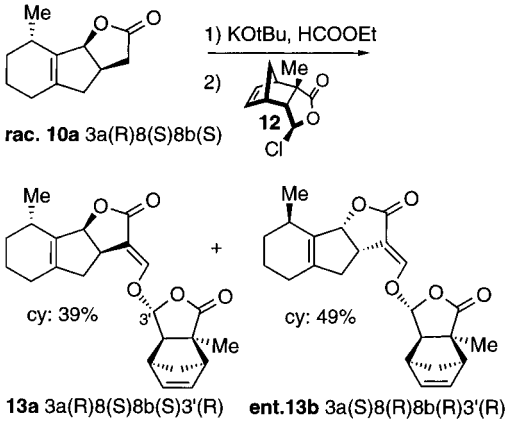


nates at the same frequency as in the natural compound, viz. 5.5 ppm, whereas in the cis-ABC diastereoisomers **rac.2c** and **rac.2d**, H8b resonates at 5.3 ppm. This comparison allows the conclusion that the original tentative assignment of a trans relationship between the A-ring methyl moiety and ring C is correct. As was known from previous syntheses of strigolactone analogues, deduction of the absolute stereochemistry at C2′ is not feasible using NMR techniques.[25] Consequently, the NMR signals that can be distinguished in Hauck's ^{1}H NMR spectra match with both diastereomers **rac.2a** and **rac.2b.** Circular dichroism spectrometry is necessary for the determination of the absolute configuration at C2′.

Recently we prepared both enantiopure latent D-ring synthons **12** and its enantiomer **ent.12.**[12,13] Trans tricyclic lactone **rac.10a** was coupled via its potassium enolate with chloro lactone **12** to give diastereomers **13a** and **ent.13b**, which were separated by flash chromatography (Scheme 6). Similarly, **rac.10a** was coupled with chlorolactone **ent.12** to give diastereomers **13b** and **ent.13a** (structures not shown). In the same manner, reaction of cis-ABC-lactone **rac.10b** with **12** provided **13c** and **ent.13d**, respectively (structures not shown). Identically, coupling of cis-ABC-lactone **rac.10b** with **ent.12** provided **13d** and **ent.13c**, respectively (Scheme 7). These reactions proceeded with complete exo selectivity, as judged from ^{1}H NMR analysis.

The thermal retro-Diels−Alder reaction of homochiral adducts **13a**−**d** and **ent.13a**−**d** was accomplished by

Scheme 6



Scheme 7

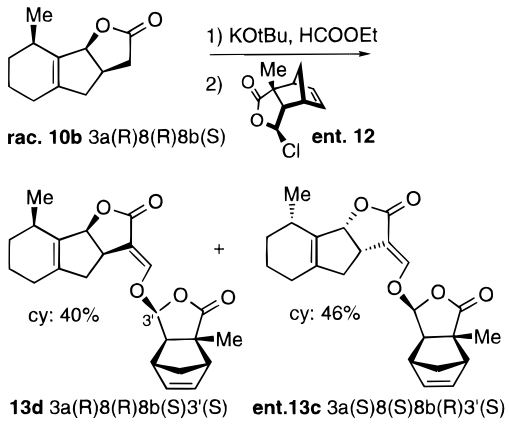
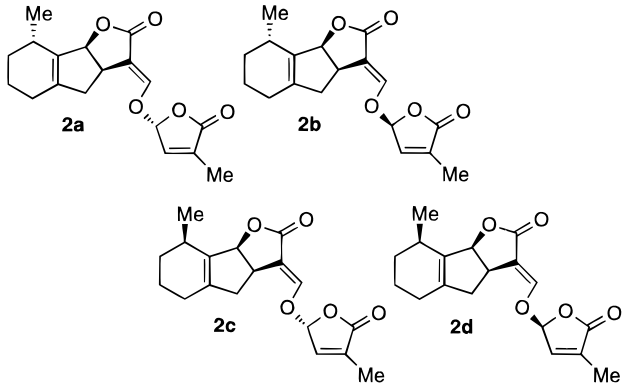


Chart 2



heating the compounds in *o*-dichlorobenzene. In this way all single sorgolactone stereoisomers **2a**−**d** and **ent.2a**−**d** were obtained (Chart 2, **ent.2a**−**d** not shown). The results obtained for the cycloreversions are collected in Table 1. It should be noted that in none of the reactions any epimerization was observed. Due to the fact that the de of tricyclic compounds **rac.10a** and **rac.10b** was not 100%, sorgolactones **2a**−**d** and **ent.2a**−**d** contained a small diastereomeric impurity, viz. their C8 epimer. These diastereomeric impurities could be largly removed by recrystallization and in the case of **ent.2a**, **2b**, and **ent.2b** by preparative HPLC.

The absolute configurations of all eight sorgolactone stereoisomers were determined from their ^{1}H NMR spectra in combination with comparison of their CD spectra (Figures 1−4) with those of the corresponding isomers of demethylsorgolactone and (+)- and (−)-

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Table 1. Cycloreversion of Adducts 13a−d and Their Enantiomers ent.13A−d

entry	adduct	product	yield (%)	purity (%)[a]	ee (%)	$[\alpha]_D$
1	**13a**	**2a**	49	>99	>99	+271.2
2	**ent.13a**	**ent.2a**	52	97.4	>99	−278.9
3	**13b**	**2b**	77	98.6	>99	+138.6
4	**ent.13b**	**ent.2b**	65	94.3	>99	−137.5
5	**13c**	**2c**	40	>99	>99	+283.4
6	**ent.13c**	**ent.2c**	46	98.9	>99	−286.5
7	**13d**	**2d**	58	96.7	>99	+170.8
8	**ent.13d**	**ent.2d**	67	97.4	>99	−167.2

[a] The impurities present were identified as diastereomers of the major products.

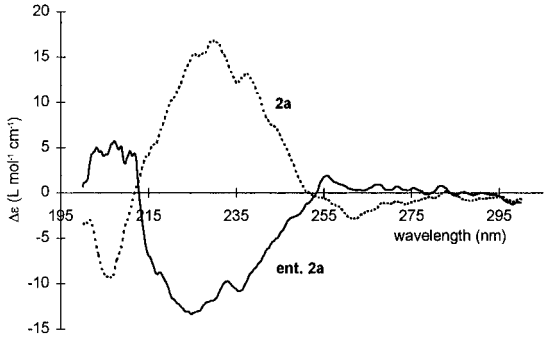

Figure 1. CD spectra of sorgolactone **2a** and its enantiomer **ent.2a**.

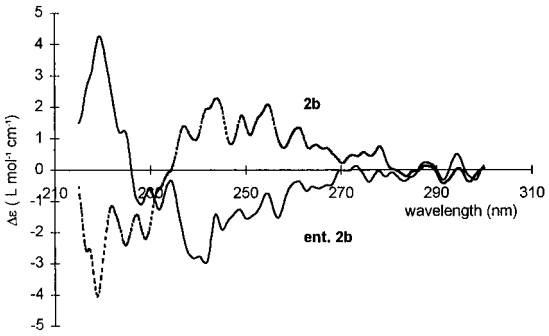

Figure 2. CD spectra of sorgolactone **2b** and its enantiomer **ent.2b**.

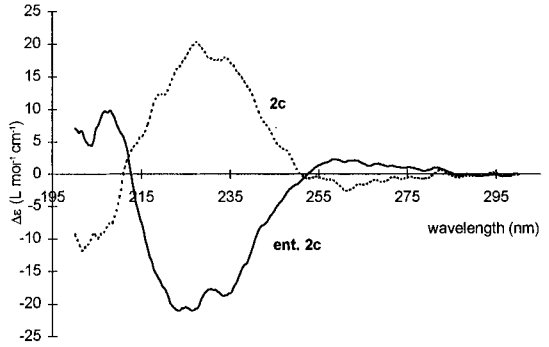

Figure 3. CD spectra of sorgolactone **2c** and its enantiomer **ent.2c**.

strigol.[15,25,26] From these previously reported data it was inferred that the sign of the Cotton effect around 270 nm could be directly correlated with the stereochemistry at C2′, a negative sign corresponding with the C2′(*R*) configuration. The configuration at C2′ as deduced from

(26) Heather, J. B.; Mittal, R. S. D.; Sih, C. J. *J. Am. Chem. Soc.* **1976**, *98*, 3661−3669.

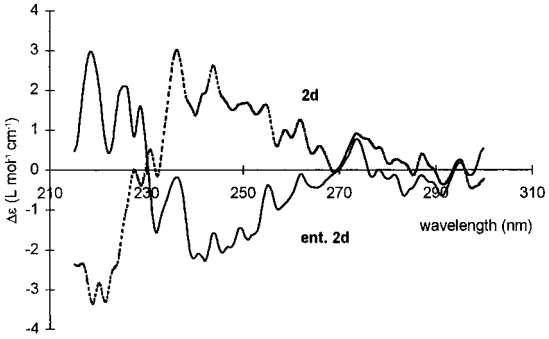

Figure 4. CD spectra of sorgolactone **2d** and its enantiomer **ent.2d**.

the CD sign around 270 nm was in all cases in complete agreement with the expected stereochemistry on the basis of the chirality of the latent D-ring synthon. Furthermore, a large Cotton effect around 240 nm in combination with a small Cotton effect of opposite sign at 270 nm proved to be indicative of a "transoid" relationship between the C ring and the stereocenter C2′ [i.e., 3a(*R*),8b(*S*),2′(*R*) and 3a(*S*),8b(*R*),2′(*S*)]. In contrast, a small Cotton effect at 240 nm and no change of sign at 270 nm corresponds with a "cisoid" relationship between ring C and C2′ [i.e., 3a(*R*),8b(*S*),2′(*S*) and 3a(*S*),8b(*R*),2′(*R*)]. A positive Cotton effect at 240 nm corresponds with the 3a(*R*),8b(*S*) configuration, whereas a negative Cotton effect at this wavelength indicates 3a(*S*),8b(*R*) stereochemistry. In this manner, all absolute configurations were unambiguously assigned. The CD spectrum of (+)-sorgolactone (**2a**) (Chart 2) was identical with that reported for the naturally occurring germination stimulant, thus proving that the proposed absolute configuration is indeed correct.

Biological Activity

The germination stimulatory activity of all eight stereoisomers of sorgolactone, **2a**−**d** and **ent.2a**−**d**, was assayed using seeds of *S. hermontica* and *O. crenata*.[27] In preliminary experiments the concentration dependent activity range (GR 24 and demethylsorgolactone) of seeds of *S. hermontica* has been established. Maximal germination percentages were obtained within the concentration range of 1 and 0.01 mg/L. Half-maximal activity was observed at approximately 0.001 mg/L. Assessment of the relative bioactivity of the individual stereoisomers of sorgolactone was therefore established at concentrations of 0.01 and 0.001 mg/L and at the sensitive concentration of 0.0001 mg/L. It was expected that the lower concentration should exhibit more profound differences. Relevant data are collected in Table 2 and represented in Figure 5.

The data presented in Table 2 reveal that there is a significant difference in germination stimulatory activity between the eight stereoisomers. For *Striga*, sorgolactone diastereomer **2a**, possessing the "natural" absolute stereochemistry, is the most active one. For *S. hermontica* the difference in activity between **2a** and most other diastereomers amounts at least a factor of 100; i.e. that the activity of about 50%, which is exhibited by all sorgolactone isomers at a concentration of 10^{-2} mg/L, is

(27) In each bioassay a diastereomeric mixture of the sorgolactone analogue GR 24 was included as a positive control and an aqueous solution of 0.1% (v/v) acetone was used as a negative control. This procedure enables a comparison between results obtained in different test series.

Table 2. Germination Percentages for Seeds of *S. Hermontica* after Exposure to Solutions (0.01, 0.001, and 0.0001 mg/L)[a] of Sorgolactone Diastereoisomers 2a−d and ent.2a−d and the Control GR 24[b]

		configuration		% germination ± SE at a concentration of		
entry	compound	at C3aC8C8b	at C2′	10^{-2} mg/L	10^{-3} mg/L	10^{-4} mg/L
1	**2a**	*RSS*	*R*	58.1 ± 6.0	60.6 ± 1.3	48.6 ± 3.7
2	**ent.2a**	*SRR*	*S*	47.6 ± 0.5	16.2 ± 5.3	7.7 ± 3.3[c]
3	**2b**	*RSS*	*S*	57.7 ± 2.1	30.5 ± 1.4	21.1 ± 5.3
4	**ent.2b**	*SRR*	*R*	40.5 ± 3.9	8.9 ± 1.9	0.2 ± 0.2[c]
5	**2c**	*RRS*	*R*	57.1 ± 4.6	50.6 ± 4.1	33.5 ± 6.9
6	**ent.2c**	*SSR*	*S*	38.4 ± 4.8	14.3 ± 3.9	1.4 ± 1.4[c]
7	**2d**	*RRS*	*S*	53.3 ± 0.8	9.2 ± 2.4	0.3 ± 0.3[c]
8	**ent.2d**	*SSR*	*R*	50.8 ± 1.1	22.9 ± 0.4	5.8 ± 1.3[c]
9	**GR 24**[a,d]	racemic		62.4 ± 2.9	55.6 ± 2.1	38.8 ± 4.9

[a] 3.16×10^{-8} mol/L, 3.16×10^{-9} mol/L, and 3.16×10^{-10} mol/L respectively for sorgolactone, 3.35×10^{-8} mol/L, 3.35×10^{-9} mol/L, and 3.35×10^{-10} mol/L, respectively, for GR 24. [b] Data presented are the mean ± SE of one representative experiment. [c] Not significantly different from aqueous control (without stimulant). [d] Equimolar mixture of two racemic diastereomers.

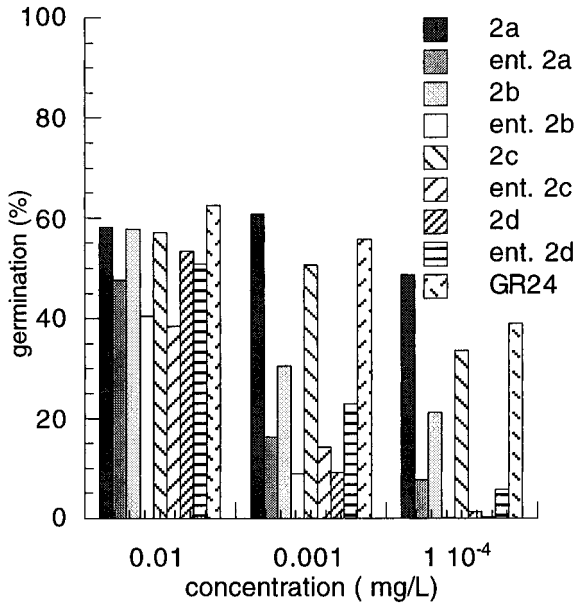

Figure 5. Bar representation of the germination percentages for seeds of *S. hermontica* after exposure to different concentrations of sorgolactone stereoisomers **2**.

at a concentration of 10^{-4} mg/L only exhibited by compound **2a**. At this sensitive concentration, the activity of almost all other sorgolactones is strongly reduced. Interestingly **2c**, which only differs from **2a** in the configuration at C8, also exhibits a relatively high germinating activity. Apparently, the absolute configuration at C8 is much less critical for the bioactivity than that of the stereocenter at C3aC8b. Remarkably, isomer **2b**, which is the C2′ epimer of "natural" sorgolactone (**2a**), also is appreciably active. The receptor for the germination stimulant still can accommodate this epimer in such a manner that induction of germination can take place. Extended bioassays involving a series of strigolactones will be needed however to allow the development of a model for the interaction of receptor and stimulant.

It is well documented[28,29] that seeds of *Orobanche* also respond to strigolactones. Therefore, for the sake of comparison, bioassays were also performed with seeds of *O. crenata*, using the same concentration range of stimulant. The results are collected in Table 3 and Figure 6. These data clearly indicate that the sorgolactones are considerably less active in germinating *Orobanche* seeds (however, the dose−response curve for *O. crenata* is evidently different from that for *S. hermontica*). As far as the absolute stereochemistry is concerned, the trend is the same as for *Striga* seeds. It should be noted that sorghum, from which sorgolactone was isolated, is not a natural host for *Orobanche crenata*.

Conclusion

We have synthesized all eight stereoisomers of the germination stimulant sorgolactone. Combination of the relative trans configuration of the natural sorgolactone ABC part (determined by NMR) with the absolute stereochemistry of the C-ring stereogenic centers C8bC3a and that of C2′ of naturally occurring sorgolactone (determined by CD spectrometry) leads to the conclusion that the proposed absolute structure of natural sorgolactone is correct. The bioactivity of all eight stereoisomers was determined, and only those isomers containing the same absolute stereochemistry as "natural" sorgolactone (**2a**) at two adjacent stereocenters exhibit significant germination stimulatory activity at sensitive concentrations.

Experimental Section

General. For general methods and instrumentation see ref 15. Final purification of sorgolactone diastereomers **ent.2a**, **2b**, and **ent.2b** was accomplished by preparative HPLC using a Lichrosorb Si60 (7 μm) column (Merck, Hibar, 250 × 25 mm) and 95/5 hexane/2-propanol as the eluent. Enantiomeric excesses and purities of stereoisomers **2a−d** and **ent.2a−d** were determined by analytical HPLC using a Chiralcel OD (10 μm) cellulose carbamate column (Baker, 250 × 4.6 mm) and 80/20 hexane/2-propanol as the eluent.

8(*S*)(*R*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*R*)(*S*)-yl)oxy]methylene]-3,3a(*R*)(*S*),4,5,6,7,8,8b(*S*)(*R*)-octahydroindeno[1,2-*b*]furan-2-one (rac.2a) and Its 2′(*S*)(*R*) Diastereomer: 8(*S*)(*R*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*S*)(*R*)-yl)oxy]methylene]-3,3a(*R*)(*S*),4,5,6,7,8,8b(*S*)(*R*)-octahydroindeno[1,2-*b*]furan-2-one (rac.2b). These products were synthesized in the same manner as **rac.2c** and **rac.2d** starting from trans tricyclic lactone **rac.10a** and chlorobutenolide **11**. Yields were 27% of fast moving diastereomer **rac.2a** and 22% of slow moving diastereomer **rac.2b**, that were both obtained as colorless crystals.

(28) Bergmann, C.; Wegmann, K.; Frischmuth, K.; Samson, E.; Kranz, A.; Weigelt, D.; Koll, P.; Welzel, P. *J. Plant Physiol.* **1993**, *142*, 338−342.

(29) Pepperman, A. B.; Dailey, O. D.; Vail, S. L. Biological activity of strigol, its precursors and its analogues. In *Synthesis and Chemistry of Agrochemicals*; Baker, D. R., Fenyes, J. G., Moberg, W. K., Cross, B., Eds.; ACS Symposium Series 355; American Chemical Society: Washington, DC, 1987; pp 445−461.

Table 3. Germination Percentages for Seeds of *O. Crenata* after Exposure to Solutions (0.1, 0.01, and 0.001 mg/L)[a] of Sorgolactone Diastereoisomers 2a−d and ent.2a−d and the Control GR 24[b]

		configuration		% germination ± SE at a concentration of		
entry	compound	at C3aC8C8b	at C2′	10^{-1} mg/L	10^{-2} mg/L	10^{-3} mg/L
1	**2a**	*RSS*	*R*	17.3 ± 0.5	5.9 ± 1.2	1.5 ± 0.8[c]
2	**ent.2a**	*SRR*	*S*	6.7 ± 1.0	1.0 ± 1.0[c]	0[c]
3	**2b**	*RSS*	*S*	10.0 ± 1.1	1.0 ± 0.5[c]	0[c]
4	**ent.2b**	*SRR*	*R*	6.4 ± 0.1	0[c]	0[c]
5	**2c**	*RRS*	*R*	17.4 ± 3.5	4.9 ± 2.9	1.7 ± 0.5
6	**ent.2c**	*SSR*	*S*	8.6 ± 0.6	1.2 ± 0.1[c]	0[c]
7	**2d**	*RRS*	*S*	8.2 ± 0.5	2.0 ± 0.1	0[c]
8	**ent.2d**	*SSR*	*R*	5.9 ± 1.9	0[c]	0[c]
9	**GR 24**[d]	racemic		27.9 ± 6.3	5.9 ± 1.0	nd[e]

[a] 3.16×10^{-7} mol/L, 3.16×10^{-8} mol/L, and 3.16×10^{-9} mol/L, respectively, for sorgolactone, 3.35×10^{-7} mol/L, 3.35×10^{-8} mol/L, and 3.35×10^{-9} mol/L, respectively, for GR 24. [b] Data presented are the mean ± SE of one representative experiment. [c] Not significantly different from aqueous control (without stimulant). [d] Equimolar mixture of two racemic diastereomers. [e] Germination percentage not determined.

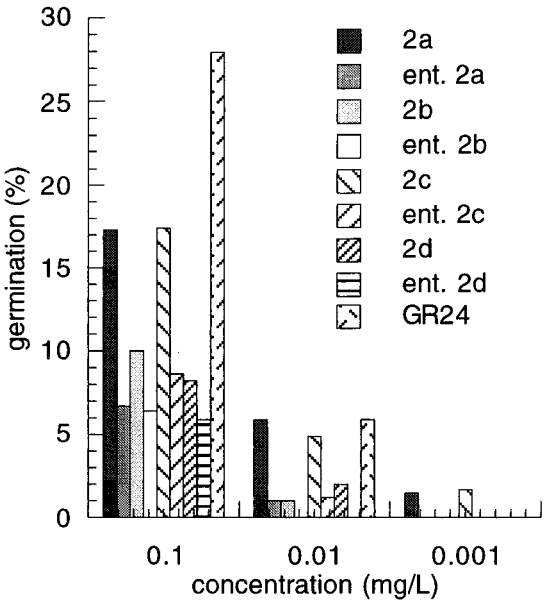

Figure 6. Bar representation of the germination percentages for seeds of *O. crenata* after exposure to different concentrations of sorgolactone stereoisomers **2**.

rac.2a: mp 114−115 °C. 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.06 (d, 3H, J = 6.9 Hz); 1.25 (m, 1H); 1.55 (m, 1H); 1.70 (m, 1H); 1.78 (m, 1H); 1.93 (m, 2H); 2.03 (s, 3H); 2.34 (bd, 1H, J = 15.9 Hz); 2.36 (m, 1H); 2.75 (dd, 1H, J = 8.5 Hz, 15.9 Hz); 3.61 (m, 1H); 5.49 (d, 1H, J = 7.6 Hz); 6.15 (s, 1H); 6.93 (s, 1H); 7.43 (d, 1H, J = 2.6 Hz). MS [EI *m*/*z*, rel intensity (%)]: 316 ($[M]^+$, 6.9); 219 ($[C_{13}H_{15}O_3]^+$, 21.8); 201 ($[C_{13}H_{13}O_2]^+$, 52.8); 173 ($[C_{12}H_{13}O]^+$, 24.7); 97 ($[C_5H_5O_2]^+$, 92.7); 91 ($[C_7H_7]^+$, 27.2). Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.25; H, 6.37.

rac.2b: mp 112−114 °C. 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.06 (d, 3H, J = 6.9 Hz); 1.25 (m, 1H); 1.55 (m, 1H); 1.70 (m, 1H); 1.77 (m, 1H); 1.93 (m, 2H); 2.03 (s, 3H); 2.33 (bd, 1H, J = 15.9 Hz); 2.36 (m, 1H); 2.73 (dd, 1H, J = 8.5 Hz, 15.9 Hz); 3.60 (m, 1H); 5.49 (d, 1H, J = 7.7 Hz); 6.14 (s, 1H); 6.94 (s, 1H); 7.43 (d, 1H, J = 2.6 Hz). MS [EI *m*/*z*, rel intensity (%)]: 316 ($[M]^+$, 8.5); 219 ($[C_{13}H_{15}O_3]^+$, 26.7); 201 ($[C_{13}H_{13}O_2]^+$, 55.7); 173 ($[C_{12}H_{13}O]^+$, 22.8); 97 ($[C_5H_5O_2]^+$, 100); 91 ($[C_7H_7]^+$, 17.0). Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.28; H, 6.36.

8(*R*)(*S*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*R*)(*S*)-yl)oxy]methylene]-3,3a(*R*)(*S*),4,5,6,7,8,8b(*S*)(*R*)-octahydroindeno[1,2-*b*]furan-2-one (rac.2c) and Its 2′(*S*)(*R*) Diastereomer 8(*R*)(*S*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*S*)(*R*)-yl)oxy]methylene]-3,3a(*R*)(*S*),4,5,6,7,8,8b(*S*)(*R*)-octahydroindeno[1,2-*b*]furan-2-one (rac.2d). To a cooled (0 °C) and stirred solution of racemic cis tricyclic lactone **rac.10b** (380 mg, 2.0 mmol) in diethyl ether (10 mL) were added, under a continuous stream of nitrogen, 3 equiv of ethyl formate (444 mg, 6.0 mmol) and 1.1 equiv of potassium *tert*-butoxide (247 mg, 2.2 mmol). The mixture was stirred overnight at room temperature. Ether was removed using a stream of nitrogen gas, and the thus obtained potassium salt of formylated **rac.10b** was dissolved in DMF (15 mL) and cooled to −50 °C. Then a solution of chlorobutenolide **11** (265 mg, 2 mmol) in DMF (3 mL) was gradually added at −50 °C under nitrogen. After overnight stirring at room temperature, the mixture was quenched with acetic acid (0.5 mL) and the solvent was removed in vacuo. The residue was dissolved in a mixture of water and ethyl acetate. The aqueous phase was extracted with ethyl acetate (2 times), and the combined organic layers were washed with brine (2 times), dried ($MgSO_4$), and concentrated under reduced pressure. The crude product was purified using flash chromatography (SiO_2, hexane/ethyl acetate 3/1) to afford two diastereomeric products. Fast moving diastereomer **rac.2c** (253 mg, 41%) and slow moving diastereomer **rac.2d** (294 mg, 47%) were obtained as colorless crystals.

rac.2c: mp 142−144 °C. 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.13 (d, 3H, J = 7.0 Hz); 1.34 (m, 1H); 1.54 (m, 1H); 1.72 (m, 2H); 1.98 (m, 2H); 2.03 (s, 3H); 2.31 (m, 1H); 2.33 (bd, 1H, J = 16.9 Hz); 2.71 (ddd, 1H, J = 3.1 Hz, 9.2 Hz, 16.8 Hz); 3.62 (m, 1H); 5.35 (d, 1H, J = 7.6 Hz); 6.15 (s, 1H); 6.92 (s, 1H); 7.42 (d, 1H, J = 2.5 Hz). MS [EI *m*/*z*, rel intensity (%)]: 316 ($[M]^+$, 9.8); 219 ($[C_{13}H_{15}O_3]^+$, 43.9); 201 ($[C_{13}H_{13}O_2]^+$, 67.7); 173 ($[C_{12}H_{13}O]^+$, 36.6); 97 ($[C_5H_5O_2]^+$, 100); 91 ($[C_7H_7]^+$, 28.7). Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.40; H, 6.37.

rac.2d: mp 138−139 °C. 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.12 (d, 3H, J = 7.0 Hz); 1.36 (m, 1H); 1.54 (m, 1H); 1.72 (m, 2H); 1.95 (m, 2H); 2.03 (s, 3H); 2.31 (m, 1H); 2.33 (bd, 1H, J = 16.9 Hz); 2.69 (ddd, 1H, J = 2.9 Hz, 12.1 Hz, 16.9 Hz); 3.61 (m, 1H); 5.35 (d, 1H, J = 7.7 Hz); 6.13 (s, 1H); 6.93 (s, 1H); 7.42 (d, 1H, J = 2.5 Hz). MS [EI *m*/*z*, rel intensity (%)]: 316 ($[M]^+$, 5.7); 219 ($[C_{13}H_{15}O_3]^+$, 43.9); 201 ($[C_{13}H_{13}O_2]^+$, 76.9); 173 ($[C_{12}H_{13}O]^+$, 40.2); 97 ($[C_5H_5O_2]^+$, 100); 91 ($[C_7H_7]^+$, 28.9). Elemental analysis was done for the enantiopure compounds, **2d** and **ent.2d**.

8(*S*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*R*)-yl)oxy]methylene]-3,3a(*R*),4,5,6,7,8,8b(*S*)-octahydroindeno[1,2-*b*]furan-2-one (2a). Deprotection of Diels−Alder adduct **13a** was accomplished by heating in *o*-dichlorobenzene as described by Thuring et al.[15] Sorgolactone stereoisomer **2a** was obtained as off-white crystals in 49% yield: mp 139.5−142 °C, $[\alpha]^{21}_D$ = +271.2° (c = 0.25, $CDCl_3$), purity 99.7%, ee >99% (determined by HPLC). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.06 (d, 3H, J = 6.9 Hz); 1.26 (m, 1H); 1.55 (m, 1H); 1.70 (m, 1H); 1.77 (m, 1H); 1.94 (m, 2H); 2.03 (s, 3H); 2.33 (bd, 1H, J = 15.4 Hz); 2.36 (m, 1H); 2.75 (dd, 1H, J = 9.0 Hz,15.4 Hz); 3.61 (m, 1H); 5.49 (d, 1H, J = 7.4 Hz); 6.15 (s, 1H); 6.92 (s, 1H); 7.41 (d, 1H, J = 2.6 Hz). ^{13}C NMR ($CDCl_3$): δ 10.8; 18.6; 20.7; 26.1; 27.8; 31.2; 36.5; 41.4; 88.0; 100.4; 114.5; 136.0; 137.3; 140.9; 141.3; 150.0; 170.2; 171.8. MS [EI *m*/*z*, rel intensity (%)]: 316 ($[M]^+$, 8.1); 219 ($[C_{13}H_{15}O_3]^+$, 25.2); 201

($[C_{13}H_{13}O_2]^+$, 48.2); 173 ($[C_{12}H_{13}O]^+$, 18.0); 97 ($[C_5H_5O_2]^+$, 100); 91 ($[C_7H_7]^+$, 14.2). Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.15; H, 6.32. CD (CH_3CN, c = 35.9μM): λ_{max} = 230 nm, $\Delta\epsilon$ = +20 L mol^{-1} cm^{-1}.

8(*R*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydro-furan-2′(*S*)-yl)oxy]methylene]-3,3a(*S*),4,5,6,7,8,8b(*R*)-octahydroindeno[1,2-*b*]furan-2-one (ent.2a). Sorgolactone stereoisomer **ent.2a** was prepared in the same manner as reported for isomer **2a** starting from **ent.13a**. **ent.2a** was obtained as a yellowish oil in 52% yield. Final purification by HPLC afforded off-white crystals: mp 145−146 °C, $[\alpha]^{21}_D$ = −278.9° (c = 0.10, CH_2Cl_2), purity 97.4%, ee >99% (determined by HPLC). ^{1}H NMR, ^{13}C NMR, and mass data were identical with those of its enantiomer **2a**. CD (CH_3CN, c = 40.5 μM): λ_{max} = 226 nm, $\Delta\epsilon$ = −13.5 L mol^{-1} cm^{-1}.

8(*S*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*S*)-yl)oxy]methylene]-3,3a(*R*),4,5,6,7,8,8b(*S*)-octahydroindeno[1,2-*b*]furan-2-one (2b). Sorgolactone stereoisomer **2b** was synthesized in the same way as described for isomer **2a** starting from **13b**. **2b** was obtained as a yellowish oil in 77% yield. Further purification by HPLC gave off-white crystals: mp 111−114 °C, $[\alpha]^{21}_D$ = +138.6° (c = 0.17, $CHCl_3$), purity 98.6%, ee >99% (determined by HPLC). ^{1}H NMR, ^{13}C NMR, and mass data were identical with those of its enantiomer **ent.2b**. CD (CH_3CN, c = 31.6μM): λ_{max} = 243 nm, $\Delta\epsilon$ = +2.5 L mol^{-1} cm^{-1}.

8(*R*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*R*)-yl)oxy]methylene]-3,3a(*S*),4,5,6,7,8,8b(*R*)-octahydroindeno[1,2-*b*]furan-2-one (ent.2b). Sorgolactone stereoisomer **ent.2b** was synthesized in the same manner as described for isomer **2a** starting from **ent.13b**. **ent.2b** was obtained as a yellowish oil in 65% yield. Chromatographic purification by HPLC afforded off-white crystals: mp 109−112 °C; $[\alpha]^{21}_D$ = −137.5° (c = 0.10, CH_2Cl_2), purity 94.3%, ee >99% (determined by HPLC). 400 MHz ^{1}H NMR ($CDCl_3$): δ 0.99 (d, 3H, J = 6.9 Hz); 1.17 (m, 1H); 1.49 (m, 1H; 1.63 (m, 1H,); 1.70 (m, 1H); 1.87 (m, 2H); 1.96 (s, 3H); 2.26 (bd, 1H, J = 15.9 Hz); 2.29 (m, 1H); 2.66 (dd, 1H, J = 8.6 Hz, 15.9 Hz); 3.53 (m, 1H); 5.43 (d, 1H, J = 7.7 Hz); 6.07 (s, 1H); 6.87 (s, 1H); 7.36 (d, 1H, J = 2.6 Hz). ^{13}C NMR ($CDCl_3$): δ 10.5; 18.6; 20.7; 26.0; 27.8; 31.6; 36.5; 41.4; 88.0; 100.6; 114.6; 135.8; 137.2; 141.0; 141.5; 150.3; 170.3; 171.8. MS [EI m/z, rel intensity (%)]: 316 ([M]$^+$, 8.9); 219 ($[C_{13}H_{15}O_3]^+$, 29.1); 201 ($[C_{13}H_{13}O_2]^+$, 53.6); 173 ($[C_{12}H_{13}O]^+$, 20.2); 97 ($[C_5H_5O_2]^+$, 96.4); 91 ($[C_7H_7]^+$, 3.1). HRMS/EI: m/z calcd for $C_{18}H_{20}O_5$ 316.13107, found 316.13098 ± 0.00088. CD (CH_3CN, c = 29.1 μM): λ_{max} = 240 nm, $\Delta\epsilon$ = −3 L mol^{-1} cm^{-1}.

8(*R*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*R*)-yl)oxy]methylene]-3,3a(*R*),4,5,6,7,8,8b(*S*)-octahydroindeno[1,2-*b*]furan-2-one (2c). Sorgolactone stereoisomer **2c** was prepared in the same manner as described for isomer **2a** starting from adduct **13c**. **2c** was obtained as colorless crystals in 40% yield: mp 178−181 °C, $[\alpha]^{21}_D$ = +284.4° (c = 0.21, $CHCl_3$), purity >99%, ee >99% (determined by HPLC). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.13 (d, 3H, J = 7.0 Hz); 1.34 (m, 1H); 1.54 (m, 1H); 1.72 (m, 2H); 1.98 (m, 2H); 2.03 (s, 3H); 2.31 (m, 1H); 2.33 (bd, 1H, J = 16.9 Hz); 2.71 (ddd, 1H, J = 3.1 Hz, 9.2 Hz, 16.8 Hz); 3.62 (m, 1H); 5.35 (d, 1H, J = 7.6 Hz); 6.15 (s, 1H); 6.92 (s, 1H); 7.42 (d, 1H, J = 2.5 Hz). ^{13}C NMR ($CDCl_3$): δ 10.8; 20.0; 20.3; 26.2; 30.1; 31.6; 36.9; 41.4; 90.9; 100.4; 114.6; 136.0; 136.6; 140.9; 142.8; 149.6; 170.2; 171.8. MS [EI m/z, rel intensity (%)]: 316 ([M]$^+$, 7.5); 219 ($[C_{13}H_{15}O_3]^+$, 32.6); 201 ($[C_{13}H_{13}O_2]^+$, 48.8); 173 ($[C_{12}H_{13}O]^+$, 20.7); 97 ($[C_5H_5O_2]^+$, 100); 91 ($[C_7H_7]^+$, 15.6). Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.63; H, 6.36. CD (CH_3CN, c = 34.2μM): λ_{max} = 227 nm, $\Delta\epsilon$ = +22 L mol^{-1} cm^{-1}.

8(*S*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*S*)-yl)oxy]methylene]-3,3a(*S*),4,5,6,7,8,8b(*R*)-octahydroindeno[1,2-*b*]furan-2-one (ent.2c). Sorgolactone stereoisomer **ent.2c** was synthesized in the same way as described for isomer **2a** starting from **ent.13c**. **ent.2c** was obtained as colorless crystals in 46% yield: mp 177.5−178.5 °C, $[\alpha]^{21}_D$ = −286.5° (c = 0.21, $CHCl_3$), purity 98.9%, ee >99% (determined by HPLC). ^{1}H NMR, ^{13}C NMR, and mass data were identical with those of its enantiomer **2c**. Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.26; H, 6.34. CD (CH_3CN, c = 33.2 μM): λ_{max} = 225 nm, $\Delta\epsilon$ = −23 L mol^{-1} cm^{-1}.

8(*R*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*S*)-yl)oxy]methylene]-3,3a(*R*),4,5,6,7,8,8b(*S*)-octahydroindeno[1,2-*b*]furan-2-one (2d). Sorgolactone stereoisomer **2d** was prepared in the same manner as described for isomer **2a** starting from adduct **13d**. **2d** was obtained as colorless crystals in 58% yield: mp 136−138 °C, $[\alpha]^{21}_D$ = +170.8° (c = 0.16, $CHCl_3$), purity 96.7%, ee >99% (determined by HPLC). ^{1}H NMR, ^{13}C, NMR and mass data were identical with those of its enantiomer **ent.2d**. Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.52; H, 6.31. CD (CH_3CN, c = 33.2 μM): λ_{max} = 243 nm, $\Delta\epsilon$ = +3 L mol^{-1} cm^{-1}.

8(*S*)-Methyl-3-[[(4′-methyl-5′-oxo-2′,5′-dihydrofuran-2′(*R*)-yl)oxy]methylene]-3,3a(*S*),4,5,6,7,8,8b(*R*)-octahydroindeno[1,2-*b*]furan-2-one (ent.2d). Sorgolactone stereoisomer **ent.2d** was synthesized in the same manner as reported for isomer **2a** starting from **ent.13d**. **ent.2d** was obtained as colorless crystals in 67% yield: mp 135−137 °C, $[\alpha]^{21}_D$ = −167.2° (c = 0.25, $CHCl_3$), purity 97.4%, ee >99% (determined by HPLC). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.12 (d, 3H, J = 7.0 Hz); 1.36 (m, 1H); 1.54 (m, 1H); 1.72 (m, 2H); 1.95 (m, 2H); 2.03 (s, 3H); 2.31 (m, 1H); 2.33 (bd, 1H, J = 16.9 Hz); 2.69 (ddd, 1H, J = 2.9 Hz, 12.1 Hz, 16.9 Hz); 3.61 (m, 1H); 5.35 (d, 1H, J = 7.7 Hz); 6.13 (s, 1H); 6.93 (s, 1H); 7.42 (d, 1H, J = 2.5 Hz). ^{13}C NMR ($CDCl_3$): δ 10.8; 20.0; 20.3; 26.2; 30.2; 31.7; 36.8; 41.5; 90.9; 100.6; 114.6; 135.9; 136.5; 141.0; 143.0; 149.9; 170.3; 171.9. MS [EI m/z, rel intensity (%)]: 316 ([M]$^+$, 7.9); 219 ($[C_{13}H_{15}O_3]^+$, 32.1); 201 ($[C_{13}H_{13}O_2]^+$, 47.7); 173 ($[C_{12}H_{13}O]^+$, 20.2); 97 ($[C_5H_5O_2]^+$, 100); 91 ($[C_7H_7]^+$, 15.5). Anal. Calcd for $C_{18}H_{20}O_5$: C, 68.34; H, 6.37. Found: C, 68.63; H, 6.36. CD (CH_3CN, c = 31.0 μM): λ_{max} = 240 nm, $\Delta\epsilon$ = −2.3 L mol^{-1} cm^{-1}.

2-[(Methoxycarbonyl)methyl]-4-methyl-3-oxo-2,3,4,5,6,7-hexahydro-1*H*-indene-2-carboxylic Acid Methyl Ester (8). Diester **8** was prepared starting from **7** analogous to the procedure described by Mangnus et al.[21] Yield: 89%. Mp: 53.5−54.5 °C. Bp: 121.5 °C (0.5 mmHg). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.09 (d, 3H, J = 7.0 Hz); 1.43 (m, 1H); 1.67−1.80 (m, 3H); 2.23−2.55 (m, 3H); 2.46 and 3.25 (AB, 2H, ^2J = 17.1 Hz); 2.48 and 3.29 (AB, 2H, ^2J = 17.3 Hz); 3.67 and 3.69 (2 × s, 2 × 3H). ^{13}C NMR ($CDCl_3$): δ 18.4; 19.0; 26.1; 28.7; 30.1; 38.6; 41.6; 51.9; 52.9; 56.2; 139.8; 170.5; 171.6; 173.7; 202.0. MS [EI m/z, rel intensity (%)]: 280 ([M]$^+$, 40.7); 220 ($[C_{13}H_{16}O_3]^+$, 100); 192 ($[C_{12}H_{16}O_2]^+$, 47.7); 133 ($[C_{10}H_{13}]^+$, 73.6); 105 ($[C_8H_9]^+$, 19.2); 91 ($[C_7H_7]^+$, 35.6). Anal. Calcd for $C_{15}H_{20}O_5$: C, 64.27; H, 7.19. Found: C, 64.48; H, 7.11.

(4-Methyl-3-oxo-2,3,4,5,6,7-hexahydro-1*H*-inden-2-yl)acetic Acid (9). Carboxylic acid **9** was synthesized from **8** in 97% yield according to the procedure reported by Mangnus et al.[21] and obtained as 1:1 mixture of cis and trans carboxylic acids **9**. Racemic trans acid **9a** precipitated when mixture **9** was dissolved in diisopropyl ether. Recrystallization from diisopropyl ether gave acid **9a** in 87% de.

9a: mp 111−113 °C, de 87% (determined by ^{13}C NMR). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.11 (d, 3H, J = 7.0 Hz); 1.42 (m, 1H); 1.64−1.81 (m, 3H); 2.21−2.30 (m, 3H); 2.41 (dd, 1H, J = 7.8 Hz, 16.3 Hz); 2.52 (m, 1H); 2.74−2.88 (m, 3H). ^{13}C NMR ($CDCl_3$): δ 18.5; 19.1; 25.9; 28.8; 30.2; 35.5; 37.2; 41.5; 141.7; 173.0; 176.0; 209.3. MS [EI m/z, rel intensity (%)]: 208 ([M]$^+$, 38.1); 190 ($[C_{12}H_{14}O_2]^+$, 36.7); 163 ($[C_{11}H_{15}O]^+$, 32.9); 147 ($[C_{11}H_{15}]^+$, 22.3); 105 ($[C_8H_9]^+$, 14.1); 91 ($[C_7H_7]^+$, 23.3); 28 ([CO]$^+$, 100). Anal. Calcd for $C_{12}H_{16}O_3$: C, 69.21; H, 7.74. Found: C, 69.24; H, 7.61. The cis diastereomer of **9** could not be obtained in diastereomerically pure form.

8(*S*)(*R*)-Methyl-3,3a(*R*)(*S*),4,5,6,7,8,8b(*S*)(*R*)-octahydroindeno[1,2-*b*]furan-2-one (rac.10a). To a solution of trans carboxylic acid **9a** (2.7 g, 13 mmol) in methanol (100 mL) were added 2 equiv of $CeCl_3 \cdot 7H_2O$ (9.7 g, 26 mmol) dissolved in methanol (100 mL) and 4 equiv of $NaBH_4$ (2.0 g, 52 mmol). The reaction mixture was stirred at room temperature for 3 h, and conversion was followed by GC. When no further progress could be detected, another equivalent of $CeCl_3 \cdot 7H_2O$ (4.8 g, 13 mmol) dissolved in methanol (50 mL) and 2 equiv of $NaBH_4$ (0.98 g, 26 mmol) were added and stirring was

continued overnight. Thereafter 1.5 equiv of $NaBH_4$ was added, and the reaction was allowed to stir until all starting material had disappeared. Methanol was evaporated in vacuo, and the residue was dissolved in a mixture of diethyl ether and a 20% aqueous solution of H_2SO_4. The aqueous phase was extracted with diethyl ether (2 times), and the combined organic layers were dried ($MgSO_4$) and concentrated in vacuo. Silica gel column chromatography (hexane/ethyl acetate 6/1) yielded racemic trans tricyclic lactone **rac.10a** as a colorless oil (2.0 g, 80%): de 87%[30] (determined by NMR). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1,05 (d, 3H, J = 7.0 Hz); 1.25 (m, 1H); 1.60 (m, 1H); 1.76 (m, 2H); 1.97 (m, 2H); 2.14 (dd, 1H, J = 1.9 Hz, 16.5 Hz); 2.29 (dd, 1H, J = 5.7 Hz, 18.4 Hz); 2.36 (m, 1H); 2.71 (dd, 1H, J = 8.2 Hz, 16.6 Hz); 2.83 (dd, 1H, J = 0.7 Hz, 18.3 Hz); 3.05 (m, 1H); 5.47 (d, 1H, J = 7.5 Hz). 400 MHz 2D-NOESY NMR ($CDCl_3$): δ 5.31 (H8b) cross peak with δ 3.04 (H3a), δ 1.11 (CH_3) *not* with δ 2.32 (H8). ^{13}C NMR ($CDCl_3$): δ 18.6; 20.7; 26.1; 27.9; 31.2; 34.0; 36.6; 42.6; 89.9; 137.4; 140.9; 177.7. MS [EI *m*/*z*, rel intensity (%)]: 192 ([M]$^+$, 72.8); 148 ([$C_{11}H_{16}$]$^+$, 49.0); 133 ([$C_{10}H_{13}$]$^+$, 100); 108 ([C_8H_{12}]$^+$, 89.6); 105 ([C_8H_9]$^+$, 64.3); 91 ([C_7H_7]$^+$, 76.4).

8(*R*)(*S*)-Methyl-3,3a(*R*)(*S*),4,5,6,7,8,8b(*S*)(*R*)-octahydroindeno[1,2-*b*]furan-2-one (rac.10b). The reaction was performed as described for compound **rac.10a.** The product was obtained (starting from the 2:1 mixture of cis and trans acids **9b** (5.4 g)) as an oily mixture of cis and trans lactones **10** (4.6 g, 92%) from which cis tricyclic lactone **rac.10b** crystallized. Recrystallization from *n*-heptane provided **rac.10b** in 87% de. Mp: 54−57 °C, de 87%[28] (determined by NMR). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.11 (d, 3H, J = 7.1 Hz); 1.35 (m, 1H); 1.55 (m, 1H); 1.73 (m, 2H); 1.98 (m, 2H); 2.19 (bd, 1H, J = 16.5 Hz); 2.32 (m, 1H); 2.35 (dd, 1H, J = 4.4 Hz, 18.2 Hz); 2.61 (ddd, 1H, J = 3.0 Hz, 8.8 Hz, 16.8 Hz); 2.81 (dd, 1H, J = 10.4 Hz, 18.0 Hz); 3.04 (m, 1H); 5.31 (d, 1H, J = 7.3 Hz). 400 MHz 2D-NOESY NMR ($CDCl_3$): δ 5.31 (H8b) cross peak with δ 3.04 (H3a), δ 1.11 (CH_3) and δ 2.32 (H8). ^{13}C NMR ($CDCl_3$): δ 19.9; 20.4; 26.2; 30.1; 31.6; 34.7; 36.2; 42.4; 92.6; 136.9; 142.8; 177.8. MS [EI *m*/*z*, rel intensity (%)]: 193 ([M + 1]$^+$, 100); 148 ([$C_{11}H_{16}$]$^+$, 13.0); 133 ([$C_{10}H_{13}$]$^+$, 79.3); 105 ([C_8H_9]$^+$, 37.0); 91 ([C_7H_7]$^+$, 15.0). HRMS/EI: *m*/*z* calcd for $C_{12}H_{16}O_2$ 192.11503, found 192.11511 ± 0.00054.

8(*S*)-Methyl-3-[[(6′(*S*)-methyl-5′-oxo-4′-oxatricyclo-[5.2.1.0$^{2',6'}$]dec-8′-en-3′(*R*)-yl)oxy]methylene]-3,3a(*R*),4,5,6,7,8,8b(*S*)-octahydroindeno[1,2-*b*]furan-2-one (13a) and Its 3a(*S*),8(*R*),8b(*R*) Diastereomer (ent.13b). To a cooled (0 °C) and stirred solution of racemic trans tricyclic lactone **rac.10a** (576 mg, 3.0 mmol) in diethyl ether (15 mL) were added, under a continuous stream of nitrogen, 3 equiv of ethyl formate (666 mg, 9 mmol) and 1.1 equiv of potassium *tert*-butoxide (370 mg, 3.3 mmol). The mixture was stirred overnight at room temperature. Ether was removed by a nitrogen flow, and the potassium salt of formylated **rac.10a** was dissolved in DMF (20 mL) and cooled to −50 °C. A solution of chloro lactone **12** (596 mg, 3.0 mmol) in DMF (5 mL) was gradually added at −50 °C under nitrogen. After being stirred overnight, the mixture was quenched with acetic acid (0.5 mL) and the solvent was removed under reduced pressure. The residue was dissolved in a mixture of water and ethyl acetate. The aqueous phase was extracted with ethyl acetate (2 times), and the combined organic layers were washed with brine (2 times), dried ($MgSO_4$), and concentrated in vacuo. The crude product was purified using flash chromatography (SiO_2, hexane/ethyl acetate 4/1) to give fast moving diastereomer **13a** (444 mg, 39%) and slow moving diastereomer **ent.13b** (558 mg, 49%) as colorless crystals.

13a: mp 150−152 °C, de 93% (determined by NMR), $[\alpha]^{21}_D$ = +174.8° (c = 0.5, CH_2Cl_2). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.06 (d, 3H, J = 7.0 Hz); 1.26 (m, 1H); 1.56 (m, 4H); 1.74 (m, 4H); 1.94 (m, 2H); 2.35 (m, 2H); 2.69 (d, 1H, J = 4.1 Hz); 2.76 (dd, 1H, J = 8.8 Hz, 15.6 Hz); 2.90 (m, 1H); 3.22 (m, 1H); 3.60 (m, 1H); 5.20 (d, 1H, J < 1 Hz); 5.47 (d, 1H, J = 7.2 Hz); 6.25 (2m, 2H); 7.34 (d, 1H, J = 2.5 Hz). ^{13}C NMR ($CDCl_3$): δ 18.6; 20.7; 22.6; 26.1; 27.8; 31.1; 36.5; 41.4; 45.2; 49.8; 52.0; 53.4; 54.5; 87.8; 103.8; 113.7; 133.5; 137.5; 137.8; 141.1; 151.0; 171.9; 178.7. MS [EI *m*/*z*, rel intensity (%)]: 382 ([M]$^+$, 2.8); 316 ([$C_{18}H_{20}O_5$]$^+$, 1.6); 202 ([$C_{13}H_{14}O_2$]$^+$, 34.3); 163 ([$C_{10}H_{11}O_2$]$^+$, 89.2); 97 ([$C_5H_5O_2$]$^+$, 100); 91 ([C_7H_7]$^+$, 15.0). Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.14; H, 6.81.

ent.13b: mp 170−171 °C, de 87% (determined by NMR), $[\alpha]^{21}_D$ = −234.0° (c = 0.5, CH_2Cl_2). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.05 (d, 3H, J = 7.0 Hz); 1.22 (m, 1H); 1.55 (m, 1H); 1.57 (s, 3H); 1.68 (m, 1H); 1.71 (m, 2H); 1.75 (m, 1H); 1.93 (m, 2H); 2.29 (bd, 1H, J = 16.5 Hz); 2.36 (m, 1H); 2.70 (dd, 1H, J = 8.2 Hz, 16.5 Hz); 2.72 (d, 1H, J = 4.2 Hz); 2.88 (m, 1H); 3.21 (m, 1H); 3.58 (m, 1H); 5.19 (d, 1H, J < 1 Hz); 5.47 (d, 1H, J = 7.2 Hz); 6.25 (2m, 2H); 7.32 (d, 1H, J = 2.6 Hz). ^{13}C NMR ($CDCl_3$): δ 18.6; 20.7; 22.5; 26.0; 27.8; 31.2; 36.5; 41.1; 45.2; 49.8; 52.0; 53.4; 54.3; 87.8; 103.7; 113.8; 133.5; 137.3; 137.8; 141.3; 151.1; 171.8; 178.8. MS [EI *m*/*z*, rel intensity (%)]: 382 ([M]$^+$, 1.0); 316 ([$C_{18}H_{20}O_5$]$^+$, 1.1); 202 ([$C_{13}H_{14}O_2$]$^+$, 29.5); 163 ([$C_{10}H_{11}O_2$]$^+$, 78.2); 97 ([$C_5H_5O_2$]$^+$, 100); 91 ([C_7H_7]$^+$, 13.2). Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.16; H, 6.77.

8(*R*)-Methyl-3-[[(6′(*R*)-methyl-5′-oxo-4′-oxatricyclo-[5.2.1.0$^{2',6'}$]dec-8′-en-3′(*S*)-yl)oxy]methylene]-3,3a(*S*),4,5,6,7,8,8b(*R*)-octahydro-indeno[1,2-*b*]furan-2-one (ent.13a) and Its 3a(*R*),8(*S*),8b(*S*) Diastereomer (13b). These compounds were prepared in the same way as described for **13a** and **ent.13b**, starting from tricyclic lactone **rac.10a** and chloro lactone **ent.12**. Yields were 33% of fast moving diastereomer **ent.13a** and 39% of slow moving diastereomer **13b**.

ent.13a: mp 111−114 °C,[31] de 97% (determined by NMR), $[\alpha]^{21}_D$ = −169.6° (c = 0.5, CH_2Cl_2). ^{1}H NMR, ^{13}C NMR, and mass data were identical with those of its enantiomer **13a**. Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.40; H, 6.89.

13b: mp 172−173 °C, de 95% (determined by NMR), $[\alpha]^{21}_D$ = +232.8° (c = 0.5, CH_2Cl_2). ^{1}H NMR, ^{13}C NMR, and mass data were identical with those of its enantiomer **ent.13b**. Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.09; H, 6.80.

8(*R*)-Methyl-3-[[(6′(*S*)-methyl-5′-oxo-4′-oxatricyclo-[5.2.1.0$^{2',6'}$]dec-8′-en-3′(*R*)-yl)oxy]methylene]-3,3a(*R*),4,5,6,7,8,8b(*S*)-octahydroindeno[1,2-*b*]furan-2-one (13c) and Its 3a(*S*),8(*S*),8b(*R*) Diastereomer (ent.13d). These compounds were prepared in the same way as described for **13a** and **ent.13b**, starting from tricyclic lactone **rac.10b** and chloro lactone **12**. Yields were 28% of fast moving diastereomer **13c** and 35% of slow moving diastereomer **ent.13d**.

13c: mp 240−242 °C, de >99% (determined by NMR), $[\alpha]^{21}_D$ = +182.4° (c = 0.5, CH_2Cl_2). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.12 (d, 3H, J = 7.0 Hz); 1.34 (m, 1H); 1.54 (m, 1H, H6); 1.58 (s, 3H); 1.72 (m, 4H); 1.94 (m, 2H); 2.33 (m, 2H); 2.68 (d, 1H, J = 4.1 Hz); 2.71 (ddd, 1H, J = 3.1 Hz, 9.3 Hz, 16.8 Hz); 2.89 (m, 1H); 3.22 (m, 1H); 3.60 (m, 1H); 5.20 (d, 1H, J < 1 Hz); 5.33 (d, 1H, J = 7.7 Hz); 6.25 (2m, 2H); 7.33 (d, 1H, J = 2.4 Hz). ^{13}C NMR ($CDCl_3$): δ 19.8; 20.3; 22.6; 26.2; 30.1; 31.6; 36.9; 41.5; 45.2; 49.8; 52.0; 53.4; 54.5; 90.7; 103.8; 113.7; 133.5; 136.8; 137.8; 142.6; 150.7; 171.9; 178.8. MS [EI *m*/*z*, rel intensity (%)]: 382 ([M]$^+$, 0.8); 316 ([$C_{18}H_{20}O_5$]$^+$, 0.5); 202 ([$C_{13}H_{14}O_2$]$^+$, 28.3); 163 ([$C_{10}H_{11}O_2$]$^+$, 80.2); 97 ([$C_5H_5O_2$]$^+$, 100); 91 ([C_7H_7]$^+$, 8.2). Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.09; H, 6.80.

ent.13d: mp 217−218.5 °C, de >99% (determined by NMR), $[\alpha]^{21}_D$ = −242.8° (c = 0.5, CH_2Cl_2). 400 MHz ^{1}H NMR ($CDCl_3$): δ 1.12 (d, 3H, J = 7.0 Hz); 1.37 (m, 1H); 1.54 (m, 1H); 1.58 (s, 3H); 1.71 (m, 2H); 1.73 (m, 2H); 1.94 (m, 2H); 2.31 (m, 2H); 2.66 (ddd, 1H, J = 3.2 Hz, 9.0 Hz, 16.8 Hz); 2.73 (d, 1H, J = 4.2H); 2.90 (m, 1H); 3.22 (m, 1H); 3.60 (m, 1H); 5.20 (d, 1H, J < 1 Hz); 5.35 (d, 1H, J = 7.8 Hz); 6.25 (2m, 2H); 7.33 (d, 1H, J = 2.5 Hz). ^{13}C NMR ($CDCl_3$): δ 20.0; 20.2; 22.5;

(30) Detailed NMR analysis revealed that the de differed slightly from that reported in ref 10.

(31) The difference in melting point as compared to that of its enantiomer **13a** is presumably due to the difference in de.

26.2; 30.1; 31.6; 36.7; 41.1; 45.1; 49.8; 51.9; 53.4; 54.3; 90.7; 103.6; 113.7; 133.5; 136.5; 137.8; 142.7; 150.8; 171.8; 178.8. MS [EI *m*/*z*, rel intensity (%)]: 382 ($[M]^+$, 0.9); 316 ($[C_{18}H_{20}O_5]^+$, 0.5); 202 ($[C_{13}H_{14}O_2]^+$, 20.4); 163 ($[C_{10}H_{11}O_2]^+$, 64.1); 97 ($[C_5H_5O_2]^+$, 100); 91 ($[C_7H_7]^+$, 4.1). Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.21; H, 6.76.

8(*S*)-Methyl-3-[[(6′(*R*)-methyl-5′-oxo-4′-oxatricyclo-[5.2.1.0$^{2',6'}$]dec-8′-en-3′(*S*)-yl)oxy]methylene]-3,3a(*S*), 4,5,6,7,8,8b(*R*)-octahydroindeno[1,2-*b*]furan-2-one (ent.13c) and Its 3a(*R*),8(*R*),8b(*S*) Stereoisomer (13d). These compounds were prepared in the same way as described for **13a** and **ent.13b**, starting from tricyclic lactone **rac.10b** and chloro lactone **ent.12**. Yields were 46% of fast moving diastereomer **ent.13c** and 40% of slow moving diastereomer **13d**.

ent.13c: mp 238−240 °C, de 97% (determined by NMR), $[\alpha]^{21}_D = -187.0°$ ($c = 0.5$, CH_2Cl_2). ^{1}H NMR, ^{13}C NMR, and mass data were identical with those of its enantiomer **13c**. Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 71.69; H, 6.79.

13d: mp 217−219 °C, de 94% (determined by NMR), $[\alpha]^{21}_D = +250.2°$ ($c = 0.5$, CH_2Cl_2). ^{1}H NMR, ^{13}C NMR, and mass data were identical with those of its enantiomer **ent.13d**. Anal. Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.52; H, 6.84.

Biological Activity. (a) Seeds. Seeds of *S. hermontica* (Del.) Benth. (from *Sorghum bicolor* (L.) Moench) and *O. crenata* Forsk. (from *Vicia faba* L.) were harvested in Burkina Faso in 1994 and Egypt in 1991, respectively, and were stored in the dark at room temperature until use in germination tests. Bioassays were carried out essentially following the procedure of Mangnus et al.[32] with minor modifications.

(b) Preparation of Test Solutions. A compound to be tested was weighed out accurately to the amount of 1 mg, dissolved in 1 mL acetone p.a., and diluted with demineralized water to 100 mL. Aliquots of this stock solution were diluted further with water to obtain test solutions containing 0.1, 0.01, 0.001, and 0.0001 mg/L test compound and 0.01, 0.001, 0.0001, and 0.00001% (v/v) acetone, respectively.

(c) Bioassays. The sterilization, conditioning, and stimulation of the seeds were performed as described by Mangnus et al.[32] for *O. crenata* and as described by Kuiper[33] for *S. hermontica*. In each test series aqueous solutions with 0.1% (v/v) acetone were used as the control. Test solutions of the stimulant GR 24 (concentrations of 0.1, 0.01, 0.001, and 0.0001 mg/L) were used as references, which enables comparison among results obtained in different test series. These positive controls are important, since the response of seeds of parasitic weeds varies considerably from test to test. All tests were performed at least in duplicate, and in each test the germination percentages were determined on six disks per treatment.

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Supporting Information Available: ^{1}H NMR spectra of **rac.10a**, **rac.10b**, and **2a** (3 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

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