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Total synthesis of two 12-nordrimanes and the pharmacological active sesquiterpene hydroquinone yahazunol

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Abstract—The synthesis of two 12-nordrimanes and yahazunol was achieved via 8-oxo-12-nordrimanic acid methyl ester. The cytotoxic activity of yahazunol and seven other sesquiterpene hydroquinones and sesquiterpene quinones has been determined.

1. Introduction

The both nordrimanes (+)-(8*S*)-12-nordrimane-8,11-diacetate ((+)-**4**) and (+)-11-hydroxy-12-nordrim-9-en-8-one ((+)-**6**) have been isolated from the marine sponge *Dysidea* sp.[1] (Fig. 1). Compound (+)-**4** exhibited a weak inhibition of the bioluminescence reaction of *Photobacterium leiognathi*, a symbiotic luminous bacterium of tropical fish.[1]

The sesquiterpene hydroquinone yahazunol (**11**),[2] zonarol (**12**), zonarone (**13**), isozonarol (**14**), isozonarone (**15**),[3] cyclozonarone (**16**),[4] zonaroic acid and chromazonarol have been obtained from the brown algae *Dictyopteris undulata* Okamura.[2] The compounds **12**–**16** and chromazonarol show feeding-deterrent activity against the young abalone *Heliotis discus* hannai.[4] The sesquiterpene hydroquinones and quinones **12**–**15**, chromazonarol, zonaroic acid and dictyochromenol possess toxicity against killifish.[5]

Recently, we published the total syntheses of **11**,[6] **12**–**16**,[6,7] spongiaquinone (**17**) and hyatellaquinone (**18**)[8] starting from (−)- and (+)-albicanal (Fig. 1).

In this paper, we describe the syntheses of the 12-nordrimanes (+)-**4**, (+)-**6** and yahazunol ((−)-**11**) starting from (±)-8-oxo-12-nordrimanic acid methyl ester ((±)-**1**). Our synthesis of the building block (±)-**1** was reported earlier.[6] The marine sesquiterpene hydroquinones and sesquiterpene quinones **11**–**18** were tested for their cytotoxic activity.

Keywords: Terpenes; Phenols; Quinones.
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2. Results and discussion

According to the procedure of Furuichi et al.[9] the racemate of (±)-8-oxo-12-nordrimanic acid methyl ester ((±)-**1**) was transformed with (2*S*,3*S*)-1,4-di-*O*-benzylthreitol to the diastereomeric mixture of dioxolanes. Hydrogenolytic splitting of the benzyl groups led to the diols, which could be separated by silica gel MPLC. Hydrolysis of the both dioxolanes with 2 N sulfuric acid in MeOH yielded (+)-**1** and (−)-**1**. The keto group in position 8 of (−)-**1** was selectively reduced with $NaBH_4/CeCl_3$ to the hydroxy function in (+)-**2** (Scheme 1). In the second step the methyl ester (+)-**2** was reduced with DIBAH to the diol (+)-**3**. The one pot reduction of (−)-**1** to (+)-**3** with $NaBH_4/CeCl_3$ and DIBAH showed a better yield. Acetylation of (+)-**3** with acetyl chloride/pyridine gave (+)-(8*S*)-12-nordrimane-8,11-diacetate ((+)-**4**).

(+)-11-Hydroxy-12-nordriman-8-one ((+)-**5**) was obtained as follows.[9] The racemate of (±)-**1** reacted with (2*R*,3*R*)-2,3-butanediol to the both diastereomers which were reduced with DIBAH to the diastereomeric alcohols. These could be separated by silica gel MPLC. Hydrolysis of both dioxolanes with Nafion NR 50 led to (+)-**5** and (−)-**5**. Pyridinium chlorochromate (PCC) oxidation of (+)-**5** gave in quantitative yield (+)-11-hydroxy-12-nordrim-9-en-8-one ((+)-**6**) (Scheme 2).

The comparison of the optical rotations of (+)-(8*S*)-12-nordrimane-8,11-diacetate ((+)-**4**) ($[\alpha]_D=+72$, MeOH) and (+)-11-hydroxy-12-nordrim-9-en-8-one ((+)-**6**) ($[\alpha]_D=+9.5$, MeOH) with natural (+)-**4**[1] ($[\alpha]_D=+36$, MeOH) and (+)-**6**[1] ($[\alpha]_D=+9.6$, MeOH) led to the absolute configurations of the both nordrimanes. We assume

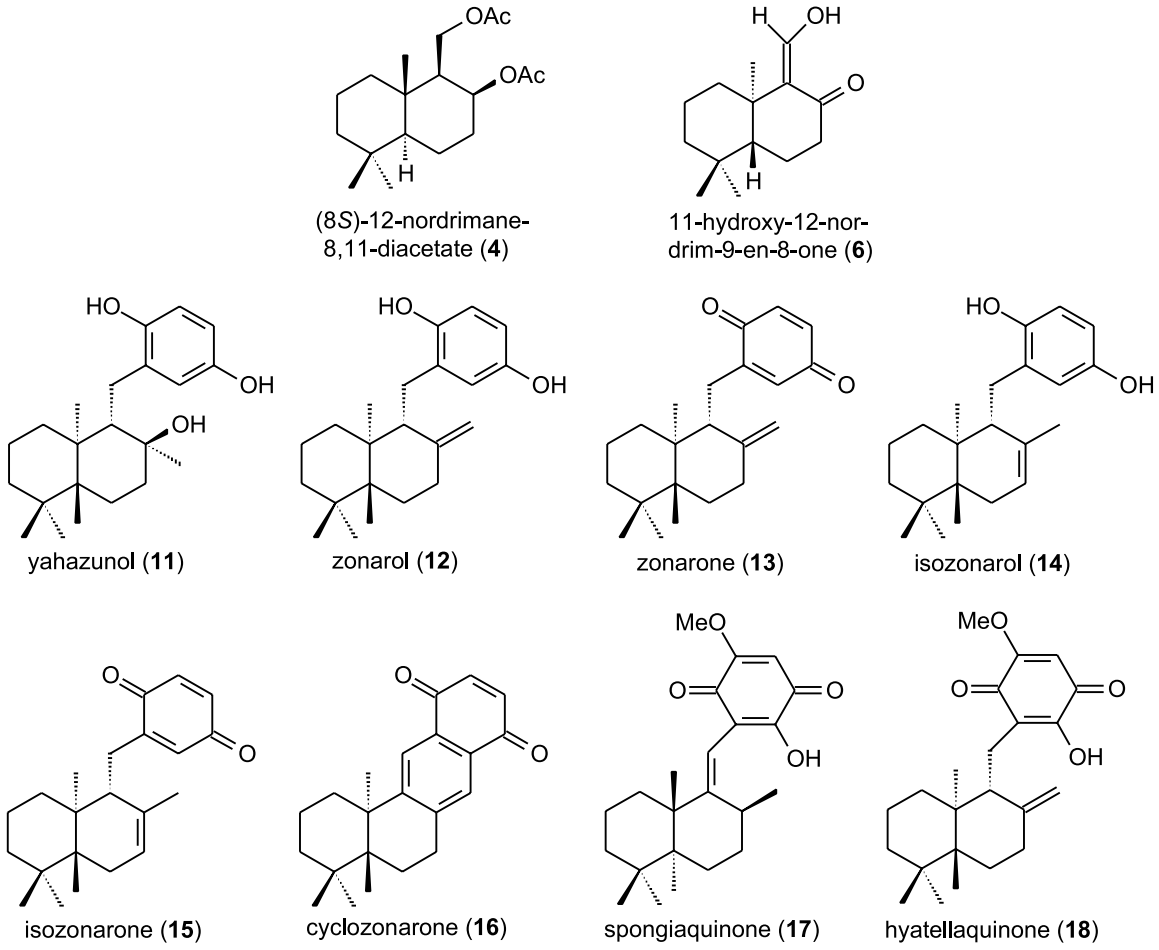
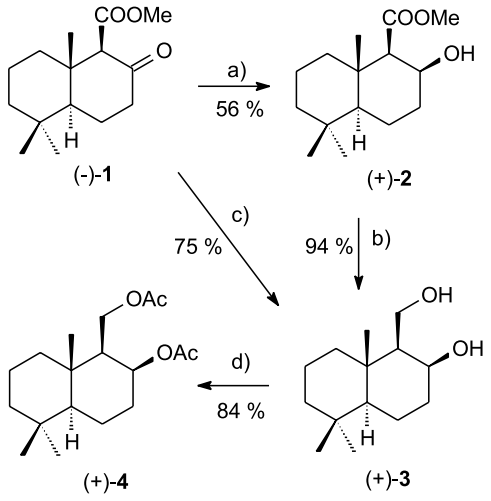


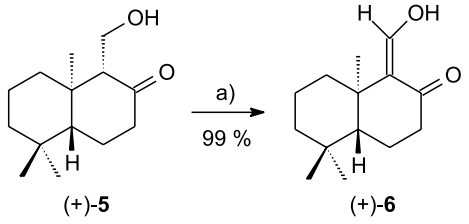
Figure 1. 12-Nordrimanes and pharmacologically tested sesquiterpene hydroquinones and sesquiterpene quinones.

that the determination of the optical rotation of natural **4** was not correct. It is interesting that the two 12-nordrimanes have been isolated from the same sponge *Dysidea* sp. but their absolute configurations are different: (+)-**4** (5*S*,10*S*), (+)-**6** (5*R*,10*R*).

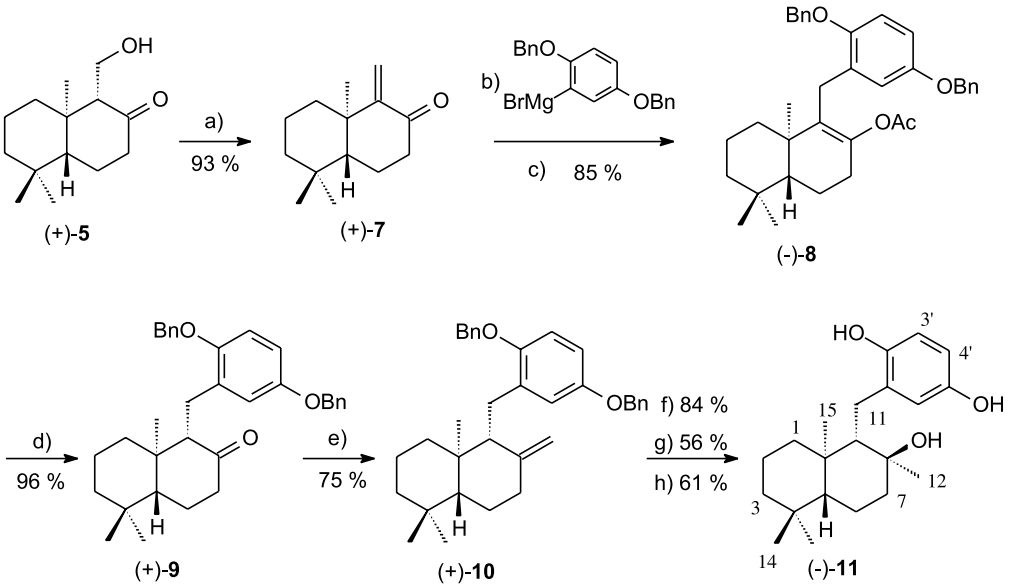


Scheme 1. Synthesis of (+)-(8*S*)-12-nordrimane-8,11-diacetat ((+)-**4**). (a) $NaBH_4$, $CeCl_3$, THF/MeOH (2:1), room temperature, 30 min; (b) DIBAH, CH_2Cl_2, room temperature, 30 min; (c) $NaBH_4$, $CeCl_3$, THF, 30 min; DIBAH, room temperature, 30 min; (d) AcCl, DMAP, Pyridin, CH_2Cl_2, room temperature, 16 h.

Our first seven step synthesis of yahazunol ((−)-**11**) started from (+)-albicanal. The change of the protecting group tetrahydopyranyl (THP) to benzyl (Bn) in (+)-**10** (Scheme 3) was a disadvantage of this route.[6] For that reason we used (+)-11-hydroxy-12-nordriman-8-one ((+)-**5**) which was transformed with *p*-toluene sulfonic acid by elimination of water to the enone (+)-**7** (Scheme 3) synthesized before starting from 12-nordrimane-8,11-diol[10,11] and (−)-**7** from 11-hydroxy-12-nordriman-8-one.[12] The cuprate catalyzed conjugated 1,4-addition[10] of 2,4-dibenzyloxyphenylmagnesium bromide to (+)-12-nordrim-9-en-8-one ((+)-**7**) yielded the enolate anion trapping with acetic anhydride. Treatment of the resulting enolacetate (−)-**8** with potassium hydroxide in methanol afforded the ketone (+)-**9**. Wittig reaction of (+)-**9** with Ph_3PCH_2 gave (+)-zonarol dibenzyl ether (+)-**10**.



Scheme 2. Synthesis of (+)-11-hydroxy-12-nordrim-9-en-8-one ((+)-**6**). (a) PCC, $CaCO_3$, CH_2Cl_2, room temperature, 20 min.



Scheme 3. Synthesis of yahazunol ((−)-**11**). (a) PTS, benzene, 50 °C, 30 min; (b) CuI, 0 °C, 10 min, room temperature, 1 h; (c) Ac_2O, 0 °C, room temperature, 20 min; (d) KOH, MeOH, room temperature, 6 h; (e) Ph_3PCH_2, THF, 80 °C, 48 h; (f) MCPBA, CH_2Cl_2, room temperature, 50 min; (g) $LiAlH_4$, Et_2O, reflux; 2 h; (h) H_2, Pd/C, EtOH, 40 °C, 30 min.

Table 1. Cytoxic activity against the tumour cell lines L-929 (murine fibroblasts), K-562 (human leukaemia) and HeLa (human cervix carcinoma) of (+)-zonarol ((+)-**12**), (+)-zonarone ((+)-**13**), (+)-isozonarol ((+)-**14**) and (+)-isozonarone ((+)-**15**)

Compound	L-929	K-562	HeLa
Zonarol ((+)-**12**)	2	2	2
Zonarone ((+)-**13**)	2	2	1
Isozonarol ((+)-**14**)	2	2	2
Isozonarone ((+)-**15**)	1	2	1

3, high activity; 2, middle activity; 1, low activity

The overall yield leading to (+)-**10** starting from (±)-8-oxo-12-nordrimanic acid methyl ester ((±)-**1**) is 24% over seven steps according to Scheme 3. The overall yield starting from (±)-**1** to (+)-**10** of our first yahazunol synthesis is 18% over 11 steps.[6] The yields of the racemate separations of (±)-**1** and (±)-albicanic acid were considered as 50%. (+)-Zonarol dibenzyl ether ((+)-**10**) was epoxidized in position 8, 12 with MCPBA. The oxirane ring was opened with $LiAlH_4$ to (+)-yahazunol dibenzyl ether as described before.[6] The yield of debenzylation of (+)-yahazunol dibenzyl ether with H_2 and Pd/C in EtOH to yahazunol ((−)-**11**) could be improved from 54 to 61% by heating the reaction mixture at 40 °C for 30 min.

The sesquiterpene hydroquinones and sesquiterpene quinones **11**–**18** (Fig. 1) were tested for their cytotoxic/cytostatic activity.

Zonarol ((+)-**12**), zonarone ((+)-**13**) and isozonarol ((+)-**14**) show a good cytotoxic activity against tumour cell lines L-929 (murine fibroblasts) and K-562 (human leukemia) and (+)-**12**, (+)-**14** a good activity against the HeLa cell line (human cervix carcinoma) (Table 1). The cytotoxic activity was determined at the Hans-Knöll-Institute in Jena according to Ref. 13.

(±)-Yahazunol ((±)-**11**), (±)-cyclozonarone ((±)-**16**), (±)-spongiaquinone ((±)-**17**) and (±)-hyatellaquinone ((±)-**18**) were investigated for their cytostatic/cytotoxic activity against the human tumour cell lines HM02 (gastric adenocarcinoma), HepG2 (hepatocellular carcinoma) and MCF 7 (breast carcinoma). (±)-Spongiaquinone ((±)-**17**) possesses the highest cytostatic/cytotoxic activity (the lowest GI50-value) against the cell lines HMO2 and

Table 2. Cytostatic/cytotoxic activity of (±)-yahazunol ((±)-**11**), (±)-cyclozonarone ((±)-**16**), (±)-spongiaquinone ((±)-**17**) and (±)-hyatellaquinone ((±)-**18**) against the tumour cell lines HM02 (gastric adenocarcinoma), HepG2 (hepatocellular carcinoma) and MCF 7 (breast carcinoma)

Compound	GI_{50}[a] (μg/ml)			TGI[b] (μg/ml)		
	HM02	HepG2	MCF 7	HM02	HepG2	MCF 7
(±)-Yahazunol (±)-**11**	4.2	7.1	6.0	10.0	>10[c]	95
(±)-Cyclozonarone (±)-**16**	5.7	9.6	>10	7.8	>10	>10
(±)-Spongiaquinone (±)-**17**	3.1	3.6	2.6	>10[d]	>10[e]	7.1
(±)-Hyatellaquinone (±)-**18**	5.3	6.0	2.4	>10[f]	>10[g]	6.9

[a] Drug concentration causing 50% growth inhibition.
[b] Drug concentration causing 100% growth inhibition.
[c] 70% inhibition at 10 μg/ml.
[d] 66% inhibition at 10 μg/ml.
[e] 52% inhibition at 10 μg/ml.
[f] 77% inhibition at 10 μg/ml.
[g] 64% inhibition at 10 μg/ml.

Table 3. Cell cycle analysis of MCF 7 cells exposed to (±)-yahazunol ((±)-**11**), (±)-spongiaquinone ((±)-**17**) and (±)-hyatellaquinone ((±)-**18**)

Compound	SubG1-phase	G1-phase	S-phase	G2/M-phase
(±)-Yahazunol ((±)-**11**) (20 μg/ml)	16.8±2.7*	44.0±3.9*	13.4±3.6	29.0±3.0*
(±)-Spongiaquinone ((±)-**17**) (40 μg/ml)	17.3±4.9*	44.0±2.4*	21.7±1.0*	16.3±1.5
(±)-Hyatellaquinone ((±)-**18**) (40 μg/ml)	23.5±3.6*	43.7±0.6*	22.2±0.4*	6.8±1.3*
Control	6.8±0.28	61.0±0.8	16.0±1.6	15.6±0.7

Data represent percentage of cells in each stage of the cell cycle. Values are mean±SE of four experiments. *$p<0.05$ versus control (*t* test).

HepG2 and hyatellaquinone ((±)-**18**) against the cell line MCF 7 (Table 2).

The cell cycle analysis of MCF 7 cells exposed to (±)-yahazunol ((±)-**11**), (±)-cyclozonarone ((±)-**16**), (±)-spongiaquinone ((±)-**17**) and (±)-hyatellaquinone ((±)-**18**) led to the following results (Table 3): (±)-yahazunol ((±)-**11**) locks the cells initially (as vincristine) in the mitose phase (G2/M-phase) and induces apoptose. (±)-Spongiaquinone ((±)-**17**) and (±)-hyatellaquinone ((±)-**18**) lock the cells initially (as 5-fluorouracil) in the synthesis phase with replication of the DNA (S-phase). This is connected with a decrease in the cell number in the G1-phase. The apoptose of the cells is based on the blockade of the DNA replication (increase in the cell number of the SubG1-phase). (±)-Cyclozonarone ((±)-**16**) was investigated in a broad concentration range. In each case, an increase in apoptotic/necrotic cells was found. A lock of the cell cycle in a specific cell phase could not be observed (data are not given). In this way (±)-yahazunol ((±)-**11**), (±)-spongiaquinone ((±)-**17**) and (±)-hyatellaquinone ((±)-**18**) block the growth of tumour cells phase specific in the G2/M- and S-phase.

3. Experimental

3.1. General

All solvents were dried and purified prior to use. THF and diethyl ether were dried by distillation from Na/K under Ar. Flash chromatography: Merck silica gel 60, 0.040–0.063 mm (230–400 mesh). MPLC: Büchi B688 pump and Büchi B687 gradient former. IR: Perkin-Elmer 1420 Ratio Recording Spectrometer; solvent $CHCl_3$. Optical rotation values: JASCO Polarimeter P-1020 (589 nm). MS: Finnigan MAT 8500 and Finnigan MAT SS 300; 70 eV.

NMR: Bruker Avance 300 and Bruker DRX 500, $CDCl_3$/$CHCl_3$, acetone-D_6/acetone and DMSO-D_6/DMSO as internal standards.

For TLC runs, precoated silica-gel foils 60 F_{254} (5×10 cm^2) from Merck were used. Spots were visualised by irradiation under UV lamp or by treatment with phosphomolybdic acid test spray.

3.2. Preparation, physical and spectroscopic data of the compounds

3.2.1. (+)-(8*S*)-8-Hydroxy-12-nordrimanic acid methyl ester ((+)-2). To a solution of $CeCl_3$ (0.4 M, 40 mmol) in THF/MeOH (100 ml, 2:1) (−)-**1** (1.00 g, 3.96 mmol) was added and stirred for 15 min. $NaBH_4$ (650 mg, 17.2 mmol) was added and stirring continued for 30 min. HCl (60 ml, 10%) and saturated NaCl-solution (20 ml) were added and the mixture was extracted three times with ethyl acetate. The combined organic layers were washed with saturated $KHCO_3$-solution and dried with Na_2SO_4. After filtration through silica gel and removing the solvent further purification was carried out by MPLC (LiChrospher® Si-60 (15 μm); hexane/ethyl acetate 7:1, 20 bar, 30 ml/min) to obtain (+)-**2** (564 mg, 2.22 mmol, 56%) as colourless crystals. Mp 164–165 °C (hexane/ethyl acetate). $[\alpha]_D^{25}=+54$ (*c* 1.00, $CHCl_3$). Ref. 10: $[\alpha]_D^{24}=+53.8$ (*c* 1.13, $CHCl_3$). IR (cm^{-1}): 3522 (w), 3010 (m), 2954 (s), 2927 (s), 2871 (m), 2850 (m), 1743 (s), 1706 (s), 1459 (m), 1438 (m), 1348 (m), 1272 (w), 1258 (w), 1232 (w), 1196 (s). MS *m*/*z* (%): 254 (17, $M^{+\cdot}$), 236 (39), 221 (94), 205 (13), 189 (10), 177 (19), 161 (61), 137 (100), 123 (95), 109 (80), 95 (85), 81 (89), 69 (87), 55 (73), 41 (89). HRMS: Calcd for $C_{15}H_{26}O_3$ 254.1882. Found 254.1882. ^1H and ^{13}C NMR: Tables 4 and 5.

3.2.2. (+)-(8*S*)-12-Nordrimane-8,11-diol ((+)-3). To a solution of (+)-**2** (470 mg, 1.85 mmol) in 10 ml of abs. CH_2Cl_2 DIBAH (Aldrich, 1.0 M in abs. CH_2Cl_2, 7.4 ml, 7.4 mmol) was added dropwise. After 30 min the solution was slowly poured into a mixture of 10 ml of concd HCl and 25 g of ice. The organic layer was separated and the aqueous layer extracted two times with ethyl acetate. The combined organic layers were washed with saturated $KHCO_3$-solution, saturated NaCl-solution and dried with Na_2SO_4. After filtration through silica gel and removing the solvent further purification was carried out by MPLC (LiChrospher® Si-60 (15 μm); hexane/ethyl acetate 2:1, 20 bar, 30 ml/min) to yield (+)-**3** (394 mg, 1.74 mmol, 94%) as colourless crystals. Mp 146–147 °C (hexane/ethyl acetate). $[\alpha]_D^{25}=+26$ (*c* 1.00, $CHCl_3$). Ref. 14: $[\alpha]_D^{23}=+26.1$ (*c* 0.93, $CHCl_3$). IR (cm^{-1}): 3500 (b), 2957 (s), 2925 (s), 2875 (m), 2847 (m), 2363 (w), 2330 (w), 1729 (m), 1457 (w), 1416 (m), 1389 (w), 1368 (w), 1334 (m), 1286 (w), 1160 (w). MS *m*/*z* (%): 226 (2, $M^{+\cdot}$), 208 (100), 193 (74), 175 (28), 165 (7), 152 (8), 149 (15), 137 (63), 123 (81), 109 (74), 95 (61), 81 (63), 69 (46), 55 (20), 41 (16). HRMS: Calcd for $C_{14}H_{26}O_2$ 226.1933. Found 226.1933. ^1H and ^{13}C NMR: Tables 4 and 5.

3.2.3. (+)-(8*S*)-12-Nordrimane-8,11-diol ((+)-3) directly from (−)-1. To a solution of $CeCl_3$ (0.4 M, 40 mmol) in THF (100 ml) (−)-**1** (1.00 g, 3.96 mmol) was added and stirred for 15 min. $NaBH_4$ (650 mg, 17.2 mmol) was added and stirring continued for 30 min. DIBAH (Aldrich, 1.0 M in abs. CH_2Cl_2, 16 ml, 16 mmol) was added dropwise. After 30 min the solution was slowly poured into a mixture of 30 ml of concd HCl and 75 g of ice. Further procedure is the same as described under Section 3.2.2 and yielded (+)-**3** (672 mg, 2.97 mmol, 75%).

Table 4. ^{1}H NMR data of compounds (+)-**2**, (+)-**3**, (+)-**4**, (+)-**6**, (+)-**7**, (−)-**8**, (+)-**9**, (+)-**10** and (−)-**11**

Position	(+)-**2**[a] ($CDCl_3$)	(+)-**3**[a] (D.-D_6)	(+)-**4**[a] ($CDCl_3$)	(+)-**6**[a] ($CDCl_3$)	(+)-**7**[a] ($CDCl_3$)	(−)-**8**[b] ($CDCl_3$)	(+)-**9**[b] ($CDCl_3$)	(+)-**10**[a] ($CDCl_3$)	(−)-**11**[a] (A.-D_6)
1	1.06	0.93	1.01 dd (13.1/3.5)	1.27	1.42	0.90	1.01	1.01	0.72 dd (17.9/4.4)
	1.31	1.67	1.69	2.01	1.75	1.61	1.61	1.73	1.84
2	1.27	1.36	1.37	1.53	1.52	1.28	1.21	1.41	1.34
	1.47	1.48	1.56	1.58	1.57	1.61	1.42	1.55	1.62
3	1.05	1.12	1.13	1.17	1.19	1.07 dd (13.2/4.4)	1.01	1.10	1.11
	1.28	1.35	1.37	1.41	1.46	1.28	1.35	1.32	1.33
5	0.82 dd (12.3/2.0)	0.82	0.88	1.14	1.38	1.19	1.34	1.10	0.94
6	1.36	1.37	1.39	1.55	1.60	1.68	1.61	1.30	1.34
	1.56	1.56	1.47	1.77	1.74	1.79	1.98	1.73	1.66
7	1.30	1.38	1.45	2.41	2.23	2.17 dd (17.0/6.2)	2.15	1.95	1.57
	1.88	1.79	1.95	2.45	2.65	2.45	2.31	2.32	1.92 dd (16.9/4.0)
8	3.99	3.53	5.10 ddd (3.4)	—	—	—	—	—	—
9	2.07 d (2.2)	0.98	1.51	—	—	—	2.49 d (9.4)	2.23	1.57
11	—	4.04	3.99 d (10.7)	8.59 d (4.4)	4.97 d (1.2)	3.31	2.69 dd (13.1/1.7)	2.73	2.40 dd (20.7/8.5)
			4.11 dd (10.7/3.8)		5.50 d (1.2)	3.33	2.85 dd (13.1/9.4)	2.81	2.85 dd (20.7/2.7)
12	—	—	—	—	—	—	—	4.63	1.30 s
								4.72	
13	0.77 s	0.83 s	0.83 s	0.89 s	0.93 s	0.91 s	0.91 s	0.86 s	0.86 s
14	0.74 s	0.79 s	0.81 s	0.83 s	0.98 s	0.82 s	0.81 s	0.80 s	0.83 s
15	1.08 s	0.91 s	0.97	1.13 s	0.89 s	1.00 s	0.73 s	0.76 s	0.97 s
1′	—	—	—	—	—	—	—	—	—
2′	—	—	—	—	—	—	—	—	—
3′	—	—	—	—	—	6.79 d (8.8)	6.78 d (8.8)	6.78 d (8.7)	6.53 d (8.5)
4′	—	—	—	—	—	6.71 dd (8.8/3.0)	6.71 dd (8.8/3.0)	6.68 dd (8.7/2.8)	6.49 dd (8.5/2.7)
5′	—	—	—	—	—	—	—	—	—
6′	—	—	—	—	—	6.81 d (3.0)	7.04 d (3.0)	6.81 d (2.8)	6.64 d (2.7)
C2′OCH_2	—	—	—	—	—	5.04	4.96	4.97	—
C5′OCH_2	—	—	—	—	—	5.02	4.99	5.01	—
Ac (8)	—	—	1.98 s	—	—	1.91 s	—		—
Ac (11)	—	—	2.00 s	—	—	—	—		—
OH	3.80 br s	3.50	—	15.37 d (4.4)	—	—	—		—
OH	—	3.97	—	—	—	—	—		—
Me	3.57 s	—	—	—	—	—	—		—
Benzyl	—	—	—	—	—	7.43—7.24	7.46—7.25	7.44—7.30	—

Coupling constants J in Hz. A.-D_6, acetone-D_6; D.-D_6, DMSO-D_6.
[a] Bruker DRX 500 spectrometer.
[b] Bruker AC 300 spectrometer.

Table 5. ^{13}C NMR data of compounds (+)-**2**, (+)-**3**, (+)-**4**, (+)-**6**, (+)-**7**, (−)-**8**, (+)-**9**, (+)-**10** and (−)-**11**

Position	(+)-**2**[a] ($CDCl_3$)	(+)-**3**[a] (D.-D_6)	(+)-**4**[a] ($CDCl_3$)	(+)-**6**[a] ($CDCl_3$)	(+)-**7**[a] ($CDCl_3$)	(−)-**8**[b] ($CDCl_3$)	(+)-**9**[b] ($CDCl_3$)	(+)-**10**[a] ($CDCl_3$)	(−)-**11**[a] (A.-D_6)
1	40.1	39.3	39.3	38.0	37.7	35.8	38.6	38.9	41.4
2	17.9	18.0	18.2	18.7	18.7	18.7	18.9	19.4	19.1
3	41.7	41.7	41.7	41.2	41.8	41.4	41.8	42.1	42.6
4	33.1	32.9	33.1	32.8	33.7	33.2	33.6	33.6	33.8
5	55.1	55.4	55.1	50.0	50.5	51.1	54.1	55.6	57.0
6	16.6	16.9	17.2	17.5	20.6	18.7	24.1	24.4	21.2
7	33.0	35.2	31.4	32.6	40.8	28.0	42.6	38.3	44.6
8	67.4	64.5	69.1	188.6	204.2	144.9	211.6	148.4	75.1
9	58.6	56.2	51.4	122.5	159.0	132.1	63.9	55.7	62.4
10	37.7	36.7	36.9	35.0	40.5	38.6	43.1	39.9	40.6
11	175.7	57.4	60.9	183.0	113.5	24.3	23.2	24.0	28.0
12	—	—	—	—	—	—	—	107.6	24.6
13	33.3	33.6	33.6	33.0	33.5	33.2	33.5	33.7	33.9
14	21.3	21.7	21.6	21.1	21.5	21.7	21.6	21.7	21.9
15	16.5	16.5	15.9	24.8	21.0	20.4	14.5	14.5	15.9
1′	—	—	—	—	—	130.7	131.8	132.5	131.2
2′	—	—	—	—	—	150.1	151.0	152.7	149.7
3′	—	—	—	—	—	112.5	112.5	112.5	117.4
4′	—	—	—	—	—	112.9	112.0	111.2	114.3
5′	—	—	—	—	—	152.8	152.5	151.1	150.4
6′	—	—	—	—	—	115.9	118.9	117.6	118.9
C2′OCH_2	—	—	—	—	—	70.8	70.9	70.9	—
C5′OCH_2	—	—	—	—	—	70.2	70.4	70.5	—
C2′Bn 1	—	—	—	—	—	137.6[c]	137.2	137.5	—
C5′Bn 1	—	—	—	—	—	137.7[c]	137.5	137.4	—
Ac (8)	—	—	21.3	—	—	20.9	—	—	—
Ac (11)	—	—	21.0	—	—	—	—	—	—
CO (8)	—	—	170.5	—	—	169.2	—	—	—
CO (11)	—	—	171.3	—	—	—	—	—	—
Me	51.0	—	—	—	—	—	—	—	—
Benzyl	—	—	—	—	—		128.4	128.5	
						127.7	128.3	128.4	
						127.6	128.2	127.8	
						127.3	128.0	127.7	
						127.2	127.7	127.6	
							127.5	127.5	—

A.-D_6, acetone-D_6; D.-D_6, DMSO-D_6.
[a] Bruker DRX 500 spectrometer.
[b] Bruker AC 300 spectrometer.
[c] Signals are exchangeable.

3.2.4. (+)-(8*S*)-12-Nordrimane-8,11-diacetate ((+)-4). To a solution of (+)-**3** (149 mg, 0.66 mmol) in abs. CH_2Cl_2 (10 ml) DMAP (20 mg), abs. pyridine (1 ml, 12 mmol) and AcCl (0.85 ml, 10 mmol) were added. After 16 h MeOH (1 ml) was added and stirring continued for 30 min. The solution was filtered through silica gel and purified using MPLC (LiChrospher® Si-60 (15 μm); hexane/ethyl acetate 6:1, 20 bar, 30 ml/min) to get (+)-**4** (172 mg, 0.55 mmol, 84%) as colourless crystals. Mp 84–85 °C (hexane/ethyl acetate). $[\alpha]_D^{24}=+72$ (*c* 1.00, MeOH). Ref. 1: $[\alpha]_D^{25}=+36.0$ (*c* 0.20, MeOH). IR (cm^{-1}): 2954 (s), 2934 (s), 2871 (m), 2850 (m), 1729 (s), 1459 (m), 1438 (w), 1390 (m), 1369 (m), 1265 (s), 1155 (w), 1125 (w), 1025 (m). MS *m*/*z* (%): 310 (2, $M^{+\cdot}$), 267 (15), 250 (8), 235 (6), 207 (61), 190 (100), 175 (76), 147 (38), 137 (49), 136 (71), 109 (54), 95 (48), 69 (36), 55 (14), 43 (56). HRMS: Calcd for $C_{18}H_{30}O_4$ 310.2144. Found 310.2144. ^1H and ^{13}C NMR: Tables 4 and 5.

3.2.5. (+)-11-Hydroxy-12-nordrim-9-en-8-one ((+)-6). Pyridinium chlorochromate (1.44 g, 6.7 mmol) and $CaCO_3$ (0.65 g, 6 mmol) were stirred for 15 min in 30 ml of abs. CH_2Cl_2. A solution of (+)-**5** (100 mg, 0.45 mmol) was added. The reaction was monitored until the educt has completely disappeared (20 min). The mixture was filtered through a short silica gel column (10 cm) to separate the chromium salts. Removing the solvent in vacuo at room temperature gave (+)-**6** (99 mg, 0.45 mmol, 99%) as colourless crystals. Mp 79–80 °C (CH_2Cl_2), $[\alpha]_D^{23}=+9.5$ (*c* 0.95, MeOH). Ref. 1: $[\alpha]_D^{25}=+9.6$ (*c* 0.10, MeOH). IR (cm^{-1}): 3500 (s), 2930 (s), 2870 (m), 1620 (s), 1590 (s), 1460 (m), 1380 (m), 1290 (w), 1200 (m), 940 (w). MS *m*/*z* (%): 222 (13, $M^{+\cdot}$), 207 (100), 189 (18), 179 (60), 137 (11), 69 (17), 41 (19). HRMS: Calcd for $C_{14}H_{22}O_2$ 222.1619. Found 222.1621. ^1H and ^{13}C NMR: Tables 4 and 5.

3.2.6. (+)-12-Nordrim-9-en-8-one ((+)-7). To a solution of (+)-**5** (400 mg, 1.78 mmol) in benzene (50 ml) *p*-toluene sulfonic acid (50 mg, 0.26 mmol) was added and the mixture was stirred for 30 min at 50 °C. Saturated Na_2CO_3-solution was added, the organic layer was separated and the aqueous layer extracted two times with ethyl acetate. The combined organic layers were washed with saturated NaCl-solution and dried with Na_2SO_4. After filtration through silica gel and removing the solvent further purification was carried out by MPLC (LiChrospher® Si-60 (15 μm); hexane/ethyl acetate 20:1, 20 bar, 30 ml/min) to obtain (+)-**7** (341 mg, 1.65 mmol, 93%) as a colourless oil. $[\alpha]_D^{24}=+70$ (*c* 1.00, $CHCl_3$). Ref. 10: $[\alpha]_D^{23}=+71.9$ (*c* 0.69, $CHCl_3$). IR (cm^{-1}): 3095 (s), 2950 (m), 2930 (m), 2870 (m), 2845 (s), 1700 (s), 1610 (m), 1460 (m), 1415 (m), 1380 (w), 1300 (w), 1280 (m), 1240 (w), 1200 (m), 1175 (m), 1100 (w), 1060 (w), 1040 (w). MS *m*/*z* (%): 206 (100, $M^{+\cdot}$), 191 (72), 69 (9), 55 (10). HRMS: Calcd for $C_{14}H_{22}O$ 206.1670. Found 206.1678. ^1H and ^{13}C NMR: Tables 4 and 5.

3.2.7. (−)-11-(2′,5′-Dibenzyloxyphenyl)-12-nordrim-8,9-en-8-yl acetate ((−)-8). A mixture of Mg (320 mg, 13 mmol) and 2-bromo-1,4-hydroquinone dibenzyl ether (4.42 g, 12 mmol) in THF (75 ml) was refluxed for 2.5 h under argon. The generated Grignard reagent was added dropwise to CuI (30 mg, 3.83 mmol) at 0 °C under argon and the whole mixture was stirred at 0 °C for 10 min. A solution of (+)-**7** (620 mg, 3.0 mmol) in THF (10 ml) was added to the above cuprate reagent, stirred for 15 min at 0 °C and then 1 h at room temperature. After cooling to 0 °C Ac_2O (2 ml, 21.8 mmol) was added to the mixture and stirring was continued for 20 min at room temperature. The reaction mixture was diluted with saturated NH_4Cl-solution and saturated $NaHCO_3$-solution, extracted with *t*-butyl methyl ether and the organic layer was separated. The aqueous layer was extracted two times with ethyl acetate, the combined organic layers were washed with saturated NaCl-solution and dried with Na_2SO_4. After filtration through silica gel and removing the solvent further purification was carried out by MPLC (LiChrospher® Si-60 (15 μm); hexane/ethyl acetate 12:1, 20 bar, 30 ml/min) to get (−)-**8** (1374 mg, 2.55 mmol, 85%) as a colourless oil. $[\alpha]_D^{23}=-4$ (*c* 0.80, $CHCl_3$). IR (cm^{-1}): 3030 (s), 2915 (m), 1747 (s), 1505 (m), 1235 (w), 1200 (m), 1020 (w). MS *m*/*z* (%): 538 (2, $M^{+\cdot}$), 495 (14), 91 (100), 43 (36). HRMS: Calcd for $C_{36}H_{42}O_4$ 538.3083. Found 538.3087. ^1H and ^{13}C NMR: Tables 4 and 5.

3.2.8. (+)-11-(2′,5′-Dibenzyloxyphenyl)-8-oxo-12-nordrimane ((+)-9). A mixture of (−)-**8** (1.8 g, 3.34 mmol) and KOH (4.0 g) in MeOH (40 ml) was stirred at room temperature for 6 h. The reaction mixture was diluted with saturated NaCl-solution, extracted with *t*-butyl methyl ether and the organic layer was separated. The aqueous layer was extracted two times with ethyl acetate, the combined organic layers were washed with saturated NaCl-solution and dried with Na_2SO_4. After filtration through silica gel and removing the solvent further purification was carried out by MPLC (LiChrospher® Si-60 (15 μm); hexane/ethyl acetate 12:1, 20 bar, 30 ml/min) to yield (+)-**9** (1593 mg, 3.21 mmol, 96%) as a colourless oil. $[\alpha]_D^{24}=+7$ (*c* 0.90, $CHCl_3$). IR (cm^{-1}): 3035 (s), 2925 (m), 1709 (s), 1485 (m), 1255 (w), 1190 (m), 1020 (w). MS *m*/*z* (%): 496 (1, $M^{+\cdot}$), 332 (10), 91 (100), 65 (13). HRMS: Calcd for $C_{34}H_{40}O_3$ 496.2977. Found 496.2979. ^1H and ^{13}C NMR: Tables 4 and 5.

3.2.9. (+)-Zonarol dibenzyl ether ((+)-10). *n*-BuLi (1.8 ml, 2.88 mmol) was added to a suspension of $Ph_3P^+MeBr^-$ (1.37 g; 5.76 mmol) in THF (40 ml) at −78 °C under argon and the mixture was stirred for 30 min. A solution of (+)-**9** (790 mg; 2.28 mmol) in THF (5 ml) was added. After stirring at 80 °C for 48 h the reaction mixture was diluted with saturated NaCl-solution (20 ml) and extracted with *t*-butyl methyl ether. The organic layer was washed with H_2O and dried with Na_2SO_4. After filtration through silica gel and removing the solvent further purification was carried out by MPLC (LiChrospher® Si-60 (15 μm); pentane/Et_2O 25:1, 20 bar, 30 ml/min) to get (+)-**10** (846 mg, 1.71 mmol, 75%) as colourless crystals. Mp 110–111 °C (pentane/Et_2O). $[\alpha]_D^{23}=+18$ (*c* 1.00, $CHCl_3$). IR (cm^{-1}): 3020 (s), 2930 (m), 1495 (m), 1227 (m), 1205 (w), 1024 (w). MS *m*/*z* (%): 494 (72, $M^{+\cdot}$), 403 (6), 213 (8), 137 (7), 123 (8), 91 (100). HRMS: Calcd for $C_{35}H_{42}O_2$ 494.3185. Found 494.3184. ^1H and ^{13}C NMR: Tables 4 and 5.

The synthesis of 8,12-epoxyzonarol dibenzyl ether, (+)-yahazunol dibenzyl ether and yahazunol ((−)-**11**) was

described before.[6] The ^{1}H and ^{13}C NMR data of (−)-**11** were given in Tables 4 and 5.

3.3. Determination of the pharmacological activity

3.3.1. Test for cytostatic/cytotoxic activity. The investigations were carried out according to the NCI guidelines with the tumour cell lines HMO2, HepG2 and MCF 7. The cells were cultivated in RPMI 1640 medium with 10% fetal bovine serum on 96-well microtiter plates. After sowing the test samples (24 h) (concentrations: 0.1, 0.5, 1.0, 5.0, 10.0 μg/ml) were added and the cells were cultivated for further 48 h. The cell number was obtained by protein determination with sulforhodanine. The test samples were dissolved in MeOH. The MeOH concentration in the test was 0.1%. The concentration activity curves led to the following values: GI50=concentration which produces a half maximum inhibition of the cell growth; TGI= concentration which produces a complete inhibition of the cell growth.

3.3.2. Cell cycle analysis. Cell cycle distribution was determined by staining DNA with propidium iodide. Cells were treated for 24 h with indicated concentrations of each drug, harvested by trypsination, washed with RPMI 1640 containing 1% fetal bovine serum and resuspended in 125 μl of a solution containing 150 μg/ml propidium iodide, 1% Triton X-100, 1% bovine serum albumin and 4 mM sodium citrate buffer, pH 7.4. After 15 min incubation at room temperature under light exclusion, the same volume of RNase A (10 μg/ml in 10 mM Tris and 15 mM NaCl, pH 7.4) was added and cells were incubated for additional 30 min at room temperature. At the end of incubation period cells were analyzed using a Becton Dickinson FACSscan and Lysis II software.

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References and notes

1. Paul, V. J.; Seo, Y.; Cho, K. W.; Rho, J.-R.; Shin, J.; Bergquist, P. R. *J. Nat. Prod.* **1997**, *60*, 1115–1120.
2. Ochi, M.; Kotsuki, H.; Muraoka, K.; Tokoroyama, T. *Bull. Chem. Soc. Jpn* **1979**, *52*, 629–630.
3. Fenical, W.; Sims, J. J.; Sqatrito, D.; Wing, R. M.; Radlick, P. *J. Org. Chem.* **1973**, *38*, 2383–2386.
4. Kurata, K.; Taniguchi, K.; Suzuki, M. *Phytochemistry* **1996**, *41*, 749–752.
5. Dave, M.-N.; Kusumi, T.; Ishitsuka, M.; Iwashita, T.; Kakisawa, H. *Heterocycles* **1984**, *22*, 2301–2307.
6. Laube, T.; Schröder, J.; Stehle, R.; Seifert, K. *Tetrahedron* **2002**, *58*, 4299–4309.
7. Schröder, J.; Matthes, B.; Seifert, K. *Tetrahedron Lett.* **2001**, *42*, 8151–8152.
8. Bernet, A.; Schröder, J.; Seifert, K. *Helv. Chim. Acta* **2003**, *86*, 2009–2020.
9. Furuichi, N.; Hata, T.; Soetjipto, H.; Kato, M.; Katsumura, S. *Tetrahedron* **2001**, *57*, 8425–8442.
10. Mori, K.; Komatsu, M. *Bull. Soc. Chim. Belg.* **1986**, *95*, 771–781.
11. Akita, H.; Nozawa, M.; Shimizu, H. *Tetrahedron: Asymmetry* **1998**, *9*, 1789–1799.
12. Pena, W.; López, J. T.; Cortés, M. *Synth. Commun.* **1989**, *16*, 2841–2850.
13. Dahse, H.-M.; Schlegel, B.; Gräfe, U. *Pharmazie* **2001**, *56*, 489–491.
14. Toshima, H.; Oikawa, H.; Toyomasu, T.; Sassa, T. *Tetrahedron* **2000**, *56*, 8443–8450.