Concise enantioselective synthesis of abscisic acid and a new analogue†

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Short and high-yielding syntheses of enantiomerically pure (*S*)-(+) and (*R*)-(−)-abscisic acid are described. The syntheses proceed through key intermediates that preferentially recrystallise as single diastereoisomers for each enantiomer. This route allows the preparation of either enantiomer of abscisic acid in *ca.* 30% overall yield, and as demonstrated, gives access to an enantiomerically pure abscisic acid analogue.

Introduction

Abscisic acid (Fig. 1) is an important phytohormone implicated in many aspects of plant growth and development. Its role in transpiration,[1] seed germination,[2] embryo maturation[3] and a number of stress responses[4] has been well characterised. The sesquiterpene exists naturally in higher plants as the dextrorotatory enantiomer **1** that has attracted considerable attention. Substantial effort has been directed towards the elucidation of its signalling pathways using affinity-based methods with enantiomerically pure abscisic acid analogues to identify binding proteins.[5] The recent discovery of an abscisic acid receptor crucially depended on the successful preparation of an active conjugate of the plant hormone and demonstrates the need for reliable methods of preparing other analogues.[6]

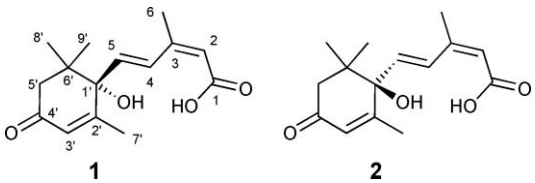


Fig. 1 The structure of (*S*)-(+)-abscisic acid (**1**) with formal atom numbering and (*R*)-(−)-abscisic acid (**2**).

Modification of the abscisic acid side-chain is an attractive approach for conjugation to supports and labels, but must be done without substantially altering either the carboxylic acid, or alkene geometry as these have been shown to be essential for biological activity.[7] In previous syntheses of abscisic acid the carbon skeleton of the side-chain has been added in one step,[8b,c,10a] limiting the opportunity to introduce the additional functionality required for our studies. Hence, following the work of Mayer[9c] and Abrams[11b] we present a new synthetic route to enantiomerically pure (*S*)-(+) and (*R*)-(−)-abscisic acid that also allows access to enantiomerically pure analogues. We demonstrate the versatility of this route by preparing a novel enantiomerically pure abscisic acid analogue that is identical to (*S*)-(+)-abscisic acid with the exception of an additional hydroxyl group on C6, Fig. 2. This new functionality will be used for conjugation of abscisic acid to other biologically useful molecules such as biotin or fluorescein without disrupting pre-existing functional groups.

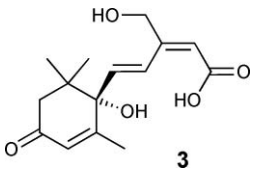
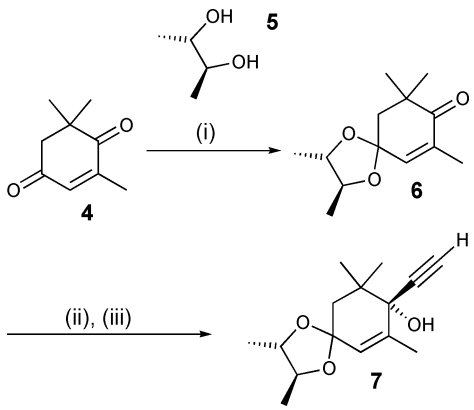


Fig. 2 (*S*)-(+)-6-Hydroxyabscisic acid (**3**).

Several formal syntheses of racemic[8] and enantiomerically pure abscisic acid[9] have been reported, however they all fail on at least one of the criteria required for efficiency, which are measured as a low number of steps, the availability of starting materials and high yields. Our new preparation of either enantiomer proceeds in *ca.* 30% overall yield in six linear steps from commercially available 4-oxoisophorone (**4**, Scheme 1), also used as a starting material for other syntheses of abscisic acid[10] and analogues.[11] Crucially, the side-chain components are easily obtained which represents a significant improvement over previous syntheses where starting materials are either no longer commercially available,[10a] or require multi-step preparation from



Scheme 1 *Reagents and conditons*: (i) **5**, TsOH, toluene, reflux 24 h (91%); (ii) LDA, trimethylsilylacetylene, THF, −78 °C to rt, 1 h, then (iii) K_2CO_3, MeOH, 2 h (71%).

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† Electronic supplementary information (ESI) available: Copies of NMR spectra for compounds **7**, **9**, **10**, **16**–**19** and **3**. ^{1}H NMR spectra and GC-MS data for compounds **7** and **14** indicating their level of diasteromeric purity. The procedure for the synthesis of (*R*)-(−)-abscisic acid including full compound characterisation data. See DOI: 10.1039/b611880a

other compounds.[9] In this new synthesis the [2(*Z*),4(*E*)] side chain is constructed stereospecifically from trimethylsilylacetylene and (*Z*)-3-iodobut-2-en-1-ol, prepared in one step by reduction of commercially available but-2-yn-1-ol with Red-Al® (sodium bis(2-methoxyethoxy)aluminium hydride), followed by iodine quench.[12] The desired stereochemistry at C1′ is obtained without chromatographic separation of diastereoisomers, thus preserving the overall efficiency of this synthesis and making it an attractive alternative to other syntheses.

Results and discussion

Previous work has seen the less hindered carbonyl of 4-oxoisophorone (**4**) protected with (2*S*,3*S*)-(+)-2,3-butanediol (**5**) to form the cyclic ketal **6**.[11b] Alkynylation proceeded in a diastereoselective fashion with the major product bearing the same configuration as (*S*)-(+)-abscisic acid. Additionally, 4-oxoisophorone, achirally protected at the C4′ carbonyl, has been shown to give a crystalline product after alkynylation with lithio trimethylsilylacetylene followed by deprotection.[10b] We found that when **6** was similarly processed and slowly recrystallised from petroleum ether, **7** was obtained in 71% yield as lustrous white needles that were of a single diastereoisomer (Scheme 1). X-Ray crystallography‡ showed this key intermediate **7** to have the same configuration at the new stereocentre as (*S*)-(+)-abscisic acid (Fig. 3). The initial addition proceeds with dr = 3 : 1 estimated by integration of NMR signals in the crude mixture at, for example 5.38 and 5.40 ppm. Upon a single recrystallisation from 40–60° petroleum ether, no minor diastereomer was visible either by ^{1}H NMR or more rigorously by GC-MS. The dr after one recrystallisation is hence estimated at ≥99 : 1 (see the ESI†).

Extension of the side chain by Sonogashira cross-coupling of **7** with (*Z*)-3-iodobut-2-en-1-ol (**8**) was achieved in 94% yield (Scheme 2). The attempted reduction of acetylene **9** with $LiAlH_4$ was unsuccessful, but, treatment with Red-Al® gave the required 4(*E*)-alkene with highest yields obtained when following aqueous quench, the reaction was stirred for 1 h before further work-up.

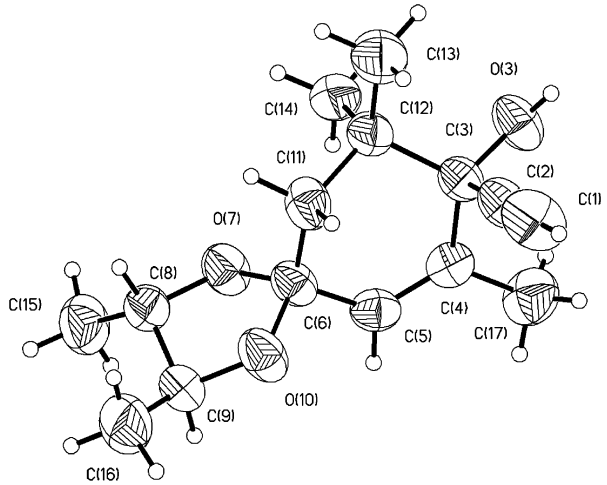
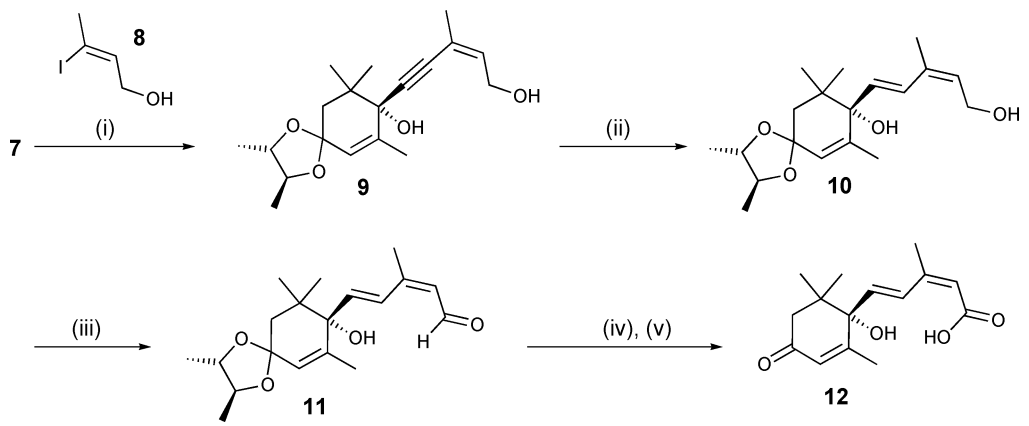


Fig. 3 The X-ray molecular structure of **7** with thermal ellipsoids at 50% probability; note crystallographic numbering used differs from Fig. 1.‡

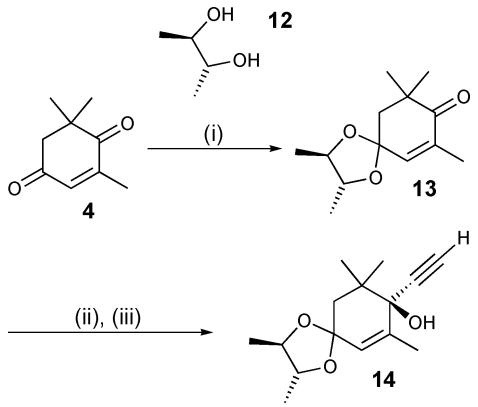
Oxidation of the primary alcohol **10** was performed cleanly in two steps *via* aldehyde **11** prepared using tetrapropylammonium perruthenate and 4-*N*-methylmorpholine-*N*-oxide as the co-oxidant,[13] followed by further oxidation using sodium chlorite with added 2-methyl-2-butene as a chlorine scavenger.[14] The cyclic ketal protecting group was removed during the acidic work-up to give (*S*)-(+)-abscisic acid (**1**) in *ca.* 30% overall yield with data identical to both natural[15] and previously synthesised material.[9a–c]

The enantiomer, (*R*)-(−)-abscisic acid (**2**) was synthesised analogously from the enantiomer of **6** by selectively protecting 4-oxoisophorone (**4**) with (2*R*,3*R*)-(−)-2,3-butanediol (**12**, Scheme 3). The resultant (2*R*,3*R*)-(−)-2,3-butanediol ketal of 4-oxoisophorone (**13**) imparts the opposite facial selectivity compared to **6** during the alkynylation with the lithio anion of trimethylsilylacetylene, giving key intermediate **14** after *in situ* removal of the trimethylsilyl protecting group. In similar fashion to the natural enantiomeric series, after a single recrystallisation a dr ≥99 : 1 (estimated from GC-MS; see the ESI†) was achieved. The trend for difference in yields (60% *vs.* 71% for the (*S*)-(+)-enantiomer) for this key C–C bond formation with concomitant deprotection was observed over several runs of this reaction. Subsequent coupling of the side chain and oxidation proceeded

‡ CCDC reference number 618045. For crystallographic data in CIF format see DOI: 10.1039/b611880a



Scheme 2 *Reagents and conditions*: (i) **8**, CuI, $PdCl_2(PPh_3)_2$, toluene, 1.5 h (94%); (ii) Red-Al®, THF, −78 °C to rt, 3 h (88%); (iii) NMO, TPAP, 4 Å mol. sieves, DCM, 20 min (95%); (iv) $NaClO_2$, KH_2PO_4, 2-methyl-2-butene, ^{t}BuOH, H_2O, 20 h, then (v) HCl (aq) (61%).



Scheme 3 *Reagents and conditions*: (i) **12**, TsOH, toluene, reflux 24 h (94%); (ii) LDA, trimethylsilylacetylene, THF, −78 °C to rt, 1 h, then (iii) K_2CO_3, MeOH, 2 h (60%).

with the same reagents and conditions, leading to (*R*)-(−)-abscisic acid (**2**, see the ESI for details†).

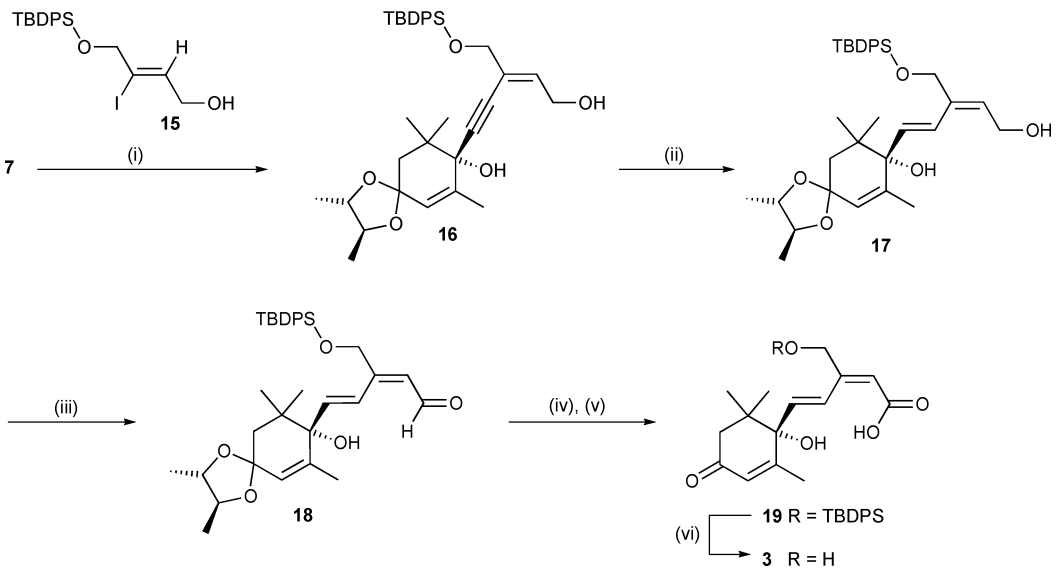
The new analogue, (*S*)-(+)-6-hydroxyabscisic acid (**3**) was prepared from the key diastereoisomerically-pure intermediate **7** and mono-protected vinyl iodide **15** (Scheme 4). Synthesis of **15** was achieved in two steps by desymmetrisation of but-2-yne-1,4-diol with *tert*-butylchlorodiphenylsilane[16] followed by reduction with Red-Al® and iodine quench.[17] Sonogashira cross-coupling of **7** with **15** was performed under a reducing atmosphere of nitrogen diluted hydrogen to reduce Hay coupling,[18] a problem not observed when preparing abscisic acid itself. Reduction of acetylene **16** with Red-Al® proceeded in low yield despite efforts at optimisation including varying reaction time, temperature and reagent stoichiometry. Incompatibility between the silyl protecting group and aluminium reducing agent appear likely since deprotection and silyl migration have been observed previously with this combination.[19] Following oxidation of the C1 alcohol and hydrolysis of the cyclic ketal the silyl protecting group was removed using tetrabutylammonium fluoride to give (*S*)-(+)-6-hydroxyabscisic acid (**3**).

Conclusion

We have developed short, efficient syntheses of enantiomerically pure (*S*)-(+) and (*R*)-(−)-abscisic acid. This enhanced route enjoys a number of improvements over previous methods including commercially available and easily prepared starting materials, and high yield of a key stereochemically defined intermediate. Uniquely, this route also allows access to new side-chain analogues that are finding application in the elucidation of abscisic acid's multiple biological activities.[20]

Experimental

Unless otherwise noted, all materials were obtained from commercial sources and used without further purification. Toluene was freshly distilled from sodium and THF from potassium. Column chromatography was performed on Merck silica gel 60 H (230–400 mesh). Melting points were recorded on a Gallenkamp melting point apparatus and are uncorrected. Optical rotations (given in 10^{-1} deg cm^2 g^{-1}) were measured on an Optical Activity AA-1000 polarimeter. IR spectra were recorded on a Perkin-Elmer Paragon 1000 FT-IR instrument. ^{1}H and ^{13}C NMR spectra were recorded using a Bruker DPX400 at 400 and 100 MHz respectively. NMR spectra were recorded in the indicated solvent and chemical shifts (δ) are reported in ppm relative to residual non-deuterated solvent as an internal standard. *J*-Values are given in Hz. Elemental analysis was performed on an Exeter Analytical CE440 machine by the Warwick Analytical Service. LSIMS and EI mass spectroscopy was performed on a Micromass Autospec mass spectrometer. GC-MS data was collected on a Varian 4000 GC-MS fitted with



Scheme 4 *Reagents and conditions*: (i) **15**, CuI, $PdCl_2(PPh_3)_2$, toluene, N_2–H_2 atm, 1 h (69%); (ii) Red-Al®, THF, −78 °C to rt, 1 h (24%); (iii) NMO, TPAP, 4 Å molecular sieves, DCM, 15 min (86%); (iv) $NaClO_2$, KH_2PO_4, 2-methyl-2-butene, ^{t}BuOH–H_2O, 20 h, then (v) HCl (aq) (68%); (vi) $Bu_4N^+F^-$, THF, 20 h (91%). TBDPS = *tert*-butyldiphenylsilane.

an autosampler and a FactorFour™ Capillary column number VF-5 ms operating a temperature programme from 50–250 °C increased at 5 °C per min.

Preparation of (*S*)-(+)-abscisic acid

(2*S*,3*S*)-2,3,7,9,9-Pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-one (6). 4-Oxoisophorone (4.22 g, 27.7 mmol, 1 eq.), 2*S*,3*S*-(+)-2,3-butanediol (3.0 g, 33.3 mmol, 1.2 eq.) and *p*-toluenesulfonic acid (160 mg) in toluene (40 ml) were heated under reflux for 24 h using a Dean–Stark trap. The mixture was cooled and washed with saturated $NaHCO_3$ solution (30 ml). The organic layer was separated and the aqueous layer extracted with diethyl ether (3 × 30 ml). The combined organics were dried over $mgSO_4$ and the solvent removed under reduced pressure. The crude product was purified by column chromatography (silica, 20% diethyl ether in hexane) to yield the title product **6** as a pale yellow oil (5.66 g, 91%). R_f 0.51 (30% diethyl ether in hexane); $[\alpha]_D^{20} = -16.1$ (*c* 0.97, MeOH) (lit.,[11b] $[\alpha]_D^{20} = -15.7$); ν_{max}(film) 1674, 1092, 969 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 1.17 (s, 3H), 1.22 (s, 3H), 1.27–1.30 (m, 6H), 1.79 (d, $J = 1.5$, 3H), 2.05 (dd, $J = 1.5$, 14.0, 1H), 2.13 (d, $J = 14.0$, 1H), 3.61–3.71 (m, 2H), 6.33 (t, $J = 1.3$, 1H); δ_C (100 MHz, $CDCl_3$) 16.2 (CH_3), 16.6 (CH_3), 16.8 (CH_3), 26.3 (CH_3), 26.8 (CH_3), 42.1 (CH_2), 47.5 (C), 78.4 (2 × CH), 102.9 (C), 135.2 (C), 141.3 (CH), 204.4 (C); MS (EI): *m*/*z* requires ($C_{13}H_{21}O_3$) 225, found 225, 100%, $[M + H]^+$.

(2*S*,3*S*,8*R*)-8-(Ethynyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro-[4.5]dec-6-en-8-ol (7). Trimethylsilylacetylene (6.14 g, 62.5 mmol, 2.5 eq.) was added slowly to a stirred solution of LDA (34.7 ml of a 1.8 M solution of LDA in THF–heptane–ethylbenzene, 2.5 eq.) in THF (200 ml) at −78 °C under a nitrogen atmosphere. The mixture was stirred for 15 min, then (2*S*,3*S*)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-one (**6**, 5.60 g, 25.0 mmol, 1 eq.) in THF (30 ml) was introduced dropwise and the mixture allowed to attain room temperature. After 1 h the reaction was quenched with saturated NH_4Cl (100 ml) and extracted with diethyl ether (3 × 100 ml). The combined organic extracts were washed with water (200 ml) then brine (200 ml) and dried over $MgSO_4$. The solvent was removed under reduced pressure to yield a pale yellow residue that was dissolved in methanol (150 ml) and treated with K_2CO_3 (20.0 g). The suspension was stirred vigorously for 2 h, then concentrated under reduced pressure. Water (50 ml) was added and the product extracted with diethyl ether (3 × 50 ml). The combined organics were washed with water (100 ml) then brine (100 ml) and dried over $MgSO_4$. The solvent was removed under reduced pressure and the residue (a mixture of diastereoisomers, 76 : 24, determined by NMR) recrystallised slowly from petroleum ether (40–60°) to give the title product **7** as white needles (4.42 g, 71%). (Found: C, 72.03; H, 8.89% $C_{15}H_{22}O_3$ requires C, 71.97; H, 8.86%); mp 140–141 °C (petroleum ether); $[\alpha]_D^{20} = +122.4$ (*c* 1.00, MeOH); ν_{max}(solid) 3427, 3285, 1090, 991, 948 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 1.08 (s, 3H), 1.16 (s, 3H), 1.23–1.26 (m, 6H), 1.82 (dd, $J = 1.2$, 14.0, 1H), 1.90 (s, 3H), 1.94 (s, 1H), 2.06 (d, $J = 14.0$, 1H), 2.49 (s, 1H), 3.53–3.63 (m, 2H), 5.38–5.39 (m, 1H); δ_C (100 MHz, $CDCl_3$) 16.7 (CH_3), 16.8 (CH_3), 18.4 (CH_3), 22.0 (CH_3), 25.5 (CH_3), 39.2 (C), 45.6 (CH_2), 74.0 (CH), 74.5 (C), 77.9 (CH), 78.0 (CH), 84.2 (C), 103.8 (C), 125.4 (CH), 140.0 (C); HRMS (EI): *m*/*z* requires ($C_{15}H_{22}O_3$) 250.1569, found 250.1563, 48%, $[M]^+$.

(2*S*,3*S*,8*R*)-8-(5-Hydroxy-3-methylpent-3-en-1-ynyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (9). Copper iodide (86 mg, 15% mol) and $PdCl_2(PPh_3)_2$ (211 mg, 10% mol), in dry deoxygenated toluene (35 ml) under a nitrogen atmosphere was treated with (*Z*)-3-iodobut-2-en-1-ol (**8**, 1.19 g, 6.0 mmol, 2 eq.) and (2*S*,3*S*,8*R*)-8-(ethynyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (**7**, 765 mg, 3.0 mmol, 1 eq.). Diisopropylamine (0.84 ml, 6.0 mmol, 2 eq.) was added dropwise and the mixture stirred for 1.5 h. Saturated NH_4Cl solution (30 ml) was added and the organic layer separated. The aqueous layer was extracted with diethyl ether (3 × 30 ml) and the combined organics dried over $MgSO_4$. The solvent was removed under reduced pressure and the product purified by column chromatography (silica, 30% EtOAc in hexane) to yield the title product **9** as a pale yellow oil (899 mg, 94%). R_f 0.19 (40% EtOAc in hexane); $[\alpha]_D^{20} = +168.2$ (*c* 1.00, MeOH) (lit.,[11b] $[\alpha]_D^{20} = +163.3$); ν_{max}(film) 3378, 1090, 982, 947 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 1.10 (s, 3H), 1.15 (s, 3H), 1.24–1.26 (m, 6H), 1.83 (dd, $J = 1.0$, 14.0, 1H), 1.86 (s, 3H), 1.91 (s, 3H), 2.04 (d, $J = 14.0$, 1H), 2.20 (s, br, 1H), 3.53–3.63 (m, 2H), 4.26 (d, $J = 6.8$, 2H), 5.37 (s, 1H), 5.86–5.89 (m, 1H); δ_C (100 MHz, $CDCl_3$) 16.7 (CH_3), 16.7 (CH_3), 18.6 (CH_3), 22.2 (CH_3), 23.0 (CH_3), 25.7 (CH_3), 39.6 (C), 45.8 (CH_2), 61.1 (CH_2), 75.0 (C), 77.9 (CH), 78.0 (CH), 83.9 (C), 94.7 (C), 103.9 (C), 120.7 (C), 125.0 (CH), 136.1 (CH), 140.4 (C); HRMS (LSIMS): *m*/*z* requires ($C_{19}H_{26}O_3$) 302.1882, found 302.1865, 84%, $[M\text{-}H_2O]^+$.

(2*S*,3*S*,8*S*)-8-(5-Hydroxy-3-methyl-2,4-pentadienyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (10). To a stirred solution of (2*S*,3*S*,8*R*)-8-(5-hydroxy-3-methylpent-3-en-1-ynyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (**9**, 635 mg, 1.9 mmol, 1 eq.) in dry deoxygenated THF (15 ml) at −78 °C under a nitrogen atmosphere was added Red-Al® (1.19 ml of a 3.2 M solution in toluene, 3.8 mmol, 2 eq.) *via* syringe. The reaction was allowed to warm to room temperature and stirred for 3 h. It was then cooled in an ice bath and water (15 ml) was carefully added dropwise and stirring continued at room temperature for 1 h. (Caution: unreacted Red-Al® reacts violently with water.) The organic layer was separated and the aqueous layer extracted with diethyl ether (3 × 15 ml). The combined organics were washed with brine (50 ml) and dried over $MgSO_4$. The solvent was removed under reduced pressure and the product purified by column chromatography (silica, 25% hexane in diethyl ether) to yield the title alcohol **10** as a viscous oil (538 mg, 88%). R_f 0.12 (25% hexane in diethyl ether); $[\alpha]_D^{20} = +199.5$ (*c* 0.75, MeOH) (lit.,[11b] $[\alpha]_D^{20} = +239.5$); ν_{max}(film) 3389, 1091, 978, 951 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 0.90 (s, 3H), 1.09 (s, 3H), 1.23 (d, $J = 5.5$, 3H), 1.26 (d, $J = 5.5$, 3H), 1.67–1.71 (m, 4H), 1.85 (d, $J = 0.7$, 3H), 1.93 (d, $J = 14.3$, 1H), 3.52–3.62 (m, 2H), 4.23–4.35 (m, 2H), 5.43 (s, 1H), 5.58 (t, $J = 7.3$, 1H), 5.72 (d, $J = 15.7$, 1H), 6.66 (d, $J = 15.7$, 1H); δ_C (100 MHz, $CDCl_3$) 16.6 (2 × CH_3), 18.0 (CH_3), 20.7 (CH_3), 23.2 (CH_3), 24.9 (CH_3), 39.3 (C), 46.3 (CH_2), 58.2 (CH_2), 77.6 (CH), 78.0 (CH), 79.4 (C), 104.0 (C), 125.5 (CH), 126.1 (CH), 128.5 (CH), 132.2 (CH), 134.8 (C),

141.6 (C); HRMS (LSIMS): *m/z* requires ($C_{19}H_{31}O_4$) 323.2222, found 323.2219, 35%, $[M + H]^+$.

(2*S*,3*S*,8*S*)-8-(3-Methylpenta-2,4-dienal)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (11). (2*S*,3*S*,8*S*)-8-(5-Hydroxy-3-methyl-2,4-pentadienyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro-[4.5]dec-6-en-8-ol (**10**, 460 mg, 1.43 mmol, 1 eq.) was dissolved in DCM (15 ml) and treated with 4-*N*-methylmorpholine-*N*-oxide (250 mg, 2.1 mmol, 1.5 eq.), powdered 4 Å molecular sieves (500 mg) and tetra-*N*-propyl ammonium perruthenate (TPAP) (50 mg, 10% mol). The mixture was stirred under nitrogen for 20 min, then filtered through a short column of silica. The column was washed with DCM and the filtrate and washings combined. The solvent was removed under reduced pressure and the product recrystallised from cyclohexane to yield the title aldehyde **11** as off-white crystals (433 mg, 95%). (Found: C, 71.22; H, 8.85%. $C_{19}H_{28}O_4$ requires C, 71.22; H, 8.81%); mp 127–128 °C (cyclohexane); $[a]_D^{20} = +348.4$ (*c* 1.14, MeOH); ν_{max}(solid) 3450, 1656, 1629, 1092, 953, 729 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 0.92 (s, 3H), 1.12 (s, 3H), 1.24 (d, $J = 5.5$, 3H), 1.27 (d, $J = 5.5$, 3H), 1.67 (d, $J = 1.5$, 3H), 1.75 (dd, $J = 1.7$, 14.6, 1H), 1.95 (d, $J = 14.6$, 1H), 2.07 (s, 3H), 3.53–3.63 (m, 2H), 5.47–5.48 (m, 1H), 5.86 (d, $J = 8.3$, 1H), 6.12 (d, $J = 15.6$, 1H), 7.35 (d, $J = 15.6$, 1H), 10.22 (d, $J = 8.3$, 1H); δ_C (100 MHz, $CDCl_3$) 16.6 (CH_3), 16.6 (CH_3), 17.8 (CH_3), 21.6 (CH_3), 23.2 (CH_3), 25.1 (CH_3), 39.4 (C), 46.3 (CH_2), 77.7 (CH), 78.1 (CH), 79.3 (C), 103.6 (C), 124.8 (CH), 126.4 (CH), 128.9 (CH), 139.5 (CH), 140.4 (C), 154.1 (C), 190.6 (CH); HRMS (LSIMS): *m/z* requires ($C_{19}H_{29}O_4$) 321.2066, found 321.2068, 100%, $[M + H]^+$.

(*S*)-5-(1-Hydroxy-2,6,6-trimethyl-4-oxo-2-cyclohexen-1-yl)-3-methyl-(2*Z*,4*E*)-pentadienoic acid (1), (*S*)-(+)-abscisic acid. A solution of KH_2PO_4 (850 mg, 6.2 mmol, 5 eq.) and sodium chlorite (1.13 g, 12.5 mmol, 10 eq.) in water (10 ml) was added dropwise over a period of 10 min to a stirred solution of (2*S*,3*S*,8*S*)-8-(3-methylpenta-2,4-dienal)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (**11**, 400 mg, 1.3 mmol, 1 eq.) and 2-methyl-2-butene (8.0 ml) in ^{t}BuOH (20 ml). The mixture was stirred for 20 h, then concentrated under reduced pressure. Methanol (10 ml) was added and the mixture carefully acidified to pH 3 with concentrated HCl (aq). After 15 min the mixture was extracted with EtOAc (3 × 20 ml) and the combined organics washed in brine (50 ml) and dried over $MgSO_4$. The solvents were removed under reduced pressure and the residue purified by column chromatography (silica, 30% EtOAc and 3% AcOH in hexane). The solid obtained was recrystallised from EtOAc–hexane to give (*S*)-(+)-abscisic acid as white crystals (200 mg, 61%). (Found: C, 67.91; H, 7.63%. $C_{15}H_{20}O_4$ requires C, 68.16; H, 7.63%); mp 159–161 °C (hexane) (lit.,[9c] mp 161–163 °C); R_f 0.27 (40% EtOAc and 3% AcOH in hexane); $[a]_D^{20} = +414.0$ (*c* 1.00, EtOH) (lit.,[9b] $[a]_D^{20} = +415.3$); ν_{max}(solid) 3394, 1646, 1622, 1597, 1248 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 1.04 (s, 3H), 1.12 (s, 3H), 1.94 (s, 3H), 2.05 (s, 3H), 2.30 (d, $J = 17.3$, 1H), 2.50 (d, $J = 17.3$, 1H), 5.77 (s, 1H), 5.98 (s, 1H), 6.18 (d, $J = 16.0$, 1H), 7.82 (d, $J = 16.0$, 1H); δ_C (100 MHz, $CDCl_3$) 19.1 (CH_3), 21.5 (CH_3), 23.1 (CH_3), 24.3 (CH_3), 41.7 (C), 49.7 (CH_2), 79.9 (C), 118.1 (CH), 127.1 (CH), 128.4 (CH), 136.9 (CH), 151.6 (C), 163.0 (C), 170.9 (C), 198.3 (C); HRMS (LSIMS): *m/z* requires ($C_{15}H_{20}O_4$) 264.1362, found 264.1362, 100%, $[M]^+$.

Preparation of (*S*)-(+)-6-hydroxyabscisic acid

(2*S*,3*S*,8*R*)-8-(5-Hydroxy-3-[*tert*-butyldiphenylsiloxymethyl]-pent-3-en-1-ynyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (16). In flame-dried glassware, (*Z*)-2-iodo-1-(*tert*-butyldiphenylsiloxy)but-2-en-4-ol (**15**, 1.6 g, 3.5 mmol, 1.1 eq.), CuI (30 mg, 5% mol) and $PdCl_2(PPh_3)_2$ (109 mg, 5% mol) were mixed in dry degassed toluene (30 ml) under a nitrogen atmosphere. $^iPr_2NH$ (1.3 ml, 9.4 mmol, 3 eq.) was added and the mixture purged with N_2–H_2 (1 : 1) for five min. (2*S*,3*S*,8*R*)-8-(ethynyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (**7**, 780 mg, 3.1 mmol, 1 eq.) in dry degassed toluene (15 ml) was added dropwise over 10 min and the mixture stirred for 1 h. Saturated NH_4Cl solution (30 ml) was added and the organic layer separated. The aqueous layer was extracted with diethyl ether (3 × 30 ml) and the combined organics dried over $MgSO_4$. The solvent was removed under reduced pressure and the product purified by column chromatography (silica, 28% EtOAc in hexane) to yield the title product **16** as a pale yellow oil (1.2 g, 69%). R_f 0.22 (40% EtOAc in hexane); $[a]_D^{20} = +97.9$ (*c* 1.25, MeOH); ν_{max}(neat) 3403, 1430, 1375, 1091, 700 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 1.06 (s, 3H), 1.06 (s, 9H), 1.11 (s, 3H), 1.21–1.25 (m, 6H), 1.74 (br, 1H), 1.80 (dd, $J = 1.1$, 14.1, 1H), 1.86 (d, $J = 1.5$, 3H), 2.01 (d, $J = 14.1$, 1H), 3.53–3.60 (m, 2H), 4.16 (d, $J = 1.5$, 2H), 4.34 (d, $J = 6.5$, 2H), 5.35 (t, $J = 1.5$, 1H), 6.27 (tt, $J = 1.8$, 6.8, 1H), 7.36–7.45 (m, 6H), 7.66 (d, $J = 7.3$, 4H); δ_C (100 MHz, $CDCl_3$) 16.7 (CH_3), 16.7 (CH_3), 18.6 (CH_3), 19.3 (C), 22.2 (3 × CH_3), 25.8 (CH_3), 26.8 (CH_3), 39.6 (C), 45.7 (CH_2), 60.9 (CH_2), 65.5 (CH_2), 75.1 (C), 77.7 (CH), 78.0 (CH), 81.2 (C), 96.1 (C), 103.9 (C), 124.2 (C), 125.1 (CH), 127.8 (4 × CH), 129.8 (2 × CH), 133.2 (2 × CH), 134.8 (CH), 135.5 (4 × CH), 140.2 (C); HRMS (LSIMS): *m/z* requires ($C_{35}H_{45}O_5Si$) 573.3026, found 573.3036, 100%, $[M–H]^+$.

(2*S*,3*S*,8*S*)-8-(5-Hydroxy-3-[*tert*-butyldiphenylsiloxymethyl]-2,4-pentadienyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (17). (2*S*,3*S*,8*R*)-8-(5-Hydroxy-3-[*tert*-butyldiphenylsiloxymethyl]pent-3-en-1-ynyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro-[4.5]dec-6-en-8-ol (**16**, 800 mg, 1.4 mmol, 1 eq.) was dissolved in anhydrous THF (80 ml) and cooled to −78 °C. Red-Al® (1.75 ml of a 3.2 M solution in toluene, 5.6 mmol, 4 eq.) was added *via* syringe and the mixture allowed to attain room temperature. After 30 min the reaction was quenched by careful addition of potassium sodium tartrate tetrahydrate (10 g) in water (40 ml). (Caution: unreacted Red-Al® reacts violently with water). The mixture was stirred for 1 h, then the organic layer separated and the aqueous layer extracted with EtOAc (3 × 50 ml). The combined organic extracts were washed with brine (100 ml), dried over $MgSO_4$ and the solvent removed under reduced pressure. The product was purified by column chromatography (silica, 30% EtOAc in hexane) to yield the title product **17** as a viscous oil (190 mg, 24%). R_f 0.26 (40% EtOAc in hexane); $[a]_D^{20} = +105.6$ (*c* 1.00, MeOH); ν_{max}(neat) 3424, 1093, 701 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 0.84 (s, 3H), 1.06 (s, 3H), 1.07 (s, 9H, ^{t}butyl), 1.21 (d, $J = 5.5$, 3H), 1.25 (d, $J = 5.2$, 3H), 1.63–1.67 (m, 4H), 1.88 (d, $J = 14.3$, 1H), 3.51–3.60 (m, 2H), 4.26–4.36 (m, 4H), 5.43 (t, $J = 1.3$, 1H), 5.67 (d, $J = 16.3$, 1H), 5.82–5.86 (m, 1H), 6.47 (d, $J = 16.0$, 1H), 7.36–7.45 (m, 6H), 7.67–7.69 (m, 4H); δ_C (100 MHz, $CDCl_3$) 16.6 (2 × CH_3), 18.1 (CH_3), 19.3 (C), 23.2 (3 × CH_3), 24.9 (CH_3), 26.9 (CH_3), 39.3 (C),

46.2 (CH_2), 58.5 (CH_2), 64.9 (CH_2), 77.6 (CH), 78.0 (CH), 79.4 (C), 103.9 (C), 123.9 (CH), 125.8 (CH), 127.6 (CH), 127.7 (4 × CH), 129.7 (2 × CH), 132.0 (CH), 133.5 (2 × C), 135.6 (4 × CH), 136.9 (C), 141.2 (C); HRMS (LSIMS): *m/z* requires ($C_{35}H_{49}O_5Si$) 577.3349, found 577.3329, 22%, $[M + H]^+$.

(2*S*,3*S*,8*S*)-8-(3-[*tert*-Butyldiphenylsiloxymethyl]penta-2,4-dienal)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (18). (2*S*,3*S*,8*S*)-8-(5-Hydroxy-3-[*tert*-butyldiphenylsiloxymethyl]-2,4-pentadienyl)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol, (275 mg, 0.48 mmol, 1 eq.) was dissolved in DCM (15 ml) and treated with tetra-*N*-propyl ammonium perruthenate (TPAP) (17 mg, 10% mol), 4-*N*-methylmorpholine-*N*-oxide (96 mg, 0.71 mmol, 1.5 eq.) and powdered 4 Å molecular sieves (280 mg). The mixture was stirred for 15 min, then filtered through a short column of silica. The silica was washed with portions of DCM and diethyl ether and the combined filtrates concentrated under reduced pressure. The crude product was purified by column chromatography (silica, 25% EtOAc in hexane) to yield the title product **18** as a viscous oil (237 mg, 86%). R_f 0.15 (25% EtOAc in hexane); $[\alpha]_D^{20} = +141.6$ (*c* 1.00, MeOH); ν_{max}(neat) 1662, 1428, 1093, 731, 701 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 0.82 (s, 3H), 1.07 (s, 3H), 1.07 (s, 9H), 1.20–1.25 (m, 6H), 1.61 (s, 3H), 1.65–1.70 (m, 1H), 1.83 (d, $J = 14.6$, 1H), 3.50–3.59 (m, 2H), 4.41 (s, 2H), 5.43 (d, $J = 1.2$, 1H), 5.87 (d, $J = 16.1$, 1H), 6.37 (d, $J = 8.3$, 1H), 7.02 (d, $J = 15.8$, 1H), 7.37–7.46 (m, 6H), 7.64–7.66 (m, 4H), 10.21 (d, $J = 8.3$, 1H); δ_C (100 MHz, $CDCl_3$) 16.6 (CH_3), 16.6 (CH_3), 17.9 (CH_3), 19.3 (C), 23.2 (3 × CH_3), 25.0 (CH_3), 26.8 (CH_3), 39.3 (C), 46.1 (CH_2), 63.7 (CH_2), 77.6 (CH), 78.1 (CH), 79.3 (C), 103.5 (C), 122.4 (CH), 125.5 (CH), 126.6 (CH), 127.9 (4 × CH), 130.0 (2 × CH), 132.7 (2 × C), 135.5 (4 × CH), 138.5 (CH), 140.0 (C), 156.3 (C), 191.4 (C); HRMS (LSIMS): *m/z* requires ($C_{35}H_{47}O_5Si$) 575.3193, found 575.3190, 100%, $[M + H]^+$.

(*S*)-5-(1-Hydroxy-2,6,6-trimethyl-4-oxo-2-cyclohexen-1-yl)-3-(*tert*-butyldiphenylsiloxymethyl)-(2*Z*,4*E*)-pentadienoic acid (19). A mixture of KH_2PO_4 (190 mg, 1.4 mmol, 5 eq.) and sodium chlorite (253 mg, 2.8 mmol, 10 eq.) in water (7 ml) was added dropwise over a period of ten min to a stirred solution of (2*S*,3*S*,8*S*)-8-(3-[*tert*-butyldiphenylsiloxymethyl]penta-2,4-dienal)-2,3,7,9,9-pentamethyl-1,4-dioxaspiro[4.5]dec-6-en-8-ol (**18**, 160 mg, 0.28 mmol, 1 eq.) and 2-methyl-2-butene (4 ml) in ^{t}BuOH (10 ml). The mixture was stirred for 20 h, then concentrated under reduced pressure to half its initial volume. MeOH (10 ml) was added and the mixture carefully acidified to pH 2 with 4 M HCl (aq). The deprotection was monitored by thin layer chromatography (silica, 40% EtOAc, 3% AcOH, 57% hexane) and typically took 15 min, after which water (20 ml) was added and the product extracted with EtOAc (3 × 30 ml). The combined organic extracts were washed with brine (60 ml), dried over $MgSO_4$ and the solvent removed under reduced pressure. The product was purified by column chromatography (silica, 30% EtOAc, 3% AcOH, 67% hexane) to yield the title product **19** as a viscous oil (98 mg, 68%). R_f 0.15 (30% EtOAc, 3% AcOH, 67% hexane); $[\alpha]_D^{20} = +394.0$ (*c* 1.00, MeOH); ν_{max}(neat) 3499, 2960, 1654, 1427, 1228, 1110, 701 cm^{-1}; δ_H (400 MHz, $CDCl_3$) 0.92 (s, 3H), 1.06 (s, 3H), 1.07 (s, 9H), 1.87 (d, $J = 1.0$, 3H), 2.21 (d, $J = 17.1$, 1H), 2.35 (d, $J = 17.1$, 1H), 4.41 (d, $J = 1.5$, 2H), 5.92 (d, $J = 16.6$, 1H), 5.93 (s, 1H), 6.22 (s, 1H), 7.38–7.45 (m, 6H), 7.57 (d, $J = 16.6$, 1H), 7.64–7.66 (m, 4H); δ_C (100 MHz, $CDCl_3$) 19.0 (CH_3), 19.2 (C), 23.1 (3 × CH_3), 24.2 (CH_3), 26.8 (CH_3), 41.6 (CH_2), 63.9 (CH_2), 79.8 (C), 115.9 (CH), 125.8 (CH), 127.2 (CH), 127.9 (4 × CH), 130.1 (2 × CH), 132.7 (2 × C), 135.0 (CH), 135.5 (4 × CH), 152.9 (C), 162.4 (C), 171.5 (C), 198.1 (C); HRMS (LSIMS): *m/z* requires ($C_{31}H_{39}O_5Si$) 519.2567, found 519.2567, 100%, $[M + H]^+$.

(*S*)-(+)-6-Hydroxyabscisic acid or (*S*)-5-(1-hydroxy-2,6,6-trimethyl-4-oxo-2-cyclohexen-1-yl)-3-hydroxymethyl-(2*Z*,4*E*)-pentadienoic acid (3). (*S*)-5-(1-Hydroxy-2,6,6-trimethyl-4-oxo-2-cyclohexen-1-yl)-3-(*tert*-butyldiphenylsiloxymethyl)-(2*Z*,4*E*)-pentadienoic acid (**19**, 93 mg, 0.18 mmol, 1 eq.) in THF (10 ml) was treated with tetrabutylammonium fluoride (0.45 ml of a 1 M solution in THF, 0.45 mmol, 2.5 eq.) and stirred for 20 h. The solvent was removed under reduced pressure and the product separated by column chromatography (silica, 80% EtOAc, 3% AcOH, 17% hexane) to yield the title product **3** as an off-white solid (46 mg, 91%). R_f 0.30 (80% EtOAc, 3% AcOH, 17% hexane); $[\alpha]_D^{20} = +430.0$ (*c* 1.00, MeOH); ν_{max}(solid) 2964, 1684, 1639, 1605, 1230, 978 cm^{-1}, δ_H (400 MHz, $CDCl_3$) 1.06 (s, 3H), 1.10 (s, 3H), 1.97 (d, $J = 1.3$, 3H), 2.22 (d, $J = 17.1$, 1H), 2.59 (d, $J = 17.1$, 1H), 4.41 (d, $J = 1.2$, 2H), 5.96 (s, 1H), 6.18 (s, 1H), 6.24 (d, $J = 16.6$, 1H), 7.63 (d, $J = 16.6$, 1H); δ_C (100 MHz, $CDCl_3$) 19.6 (CH_3), 23.6 (CH_3), 24.7 (CH_3), 42.9 (CH_2), 50.7 (C), 63.2 (CH_2), 80.6 (C), 117.4 (CH), 127.0 (CH), 127.6 (CH), 136.4 (CH), 153.1 (C), 166.5 (C), 169.9 (C),[21] 201.1 (C); HRMS (LSIMS): *m/z* requires ($C_{15}H_{21}O_5$) 281.1389, found 281.1386, 100%, $[M + H]^+$.

Crystal data for 7‡. $C_{15}H_{22}O_3$, $M = 250.33$, colourless block, 0.60 × 0.25 × 0.01 mm, monoclinic, P2(1) (No 4), $\beta = 95.392(3)°$, $a = 7.6916(3)$, $b = 11.7602(5)$, $c = 8.2773(4)$Å, $T = 298$ K, μ(Cu Kα) = 0.154 mm^{-1}, $U = 745.41(6)$ $Å^3$, Z 2, $D_{cal} = 1.115$ g cm^{-3}, $\mu = 0.610$, 4874 reflections measured, 2247 unique [$R_{int} = 0.0301$], $R\,[I>2\sigma(I)] = 0.0370$, $wR\,[I>2\sigma(I)] = 0.0879$, $GooF = 1.062$.

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