Structural separation of biological activities of jasmonates and related compounds

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A wide range of compounds derived from the basic structure of jasmonic acid, when tested for biological activity in different bioassays (*Eschscholzia californica* elicitation, *Bryonia dioica* tendril coiling, tomato transpiration and senescence and barley senescence assays) display, in each of the assays, an activity profile that is distinctly characteristic and different between assays. While differences in uptake, metabolism and/or sequestration of the compounds may account for some of the effects observed, the data allow the conclusion that structural requirements are also different for different physiological responses regulated by jasmonates. While jasmonic acid itself is active in all assays employed, some of the compounds tested in our study display a much narrower range of biological effects. Thus, tailoring of jasmonate analogues for specific applications and lacking undesirable side effects should be possible.

Introduction

There has been increased interest in recent years in jasmonic acid and related compounds, the jasmonates, because of their diverse biological activity.[1] Of particular relevance was the finding that jasmonic acid is a single transducer in plant herbivore defence[2] and in the process of pathogen defence triggered by elicitors.[3] The pleiotropic effects of jasmonic acid,[1] however, unfortunately preclude its agronomical use as an inducer of endogenous plant defence mechanisms and thus as an agent of biological plant protection.

The molecular basis of jasmonate action is still completely unknown and systematic structure–activity studies are not available. The present study was initiated in order to determine whether or not there is a common structural basis for the different jasmonate effects on plants. The data show that (a) structurally related jasmonate analogues may, nevertheless, display drastically different profiles of biological activity and (b) that unwanted side effects can be reduced or even eliminated, whilst desirable biological characteristics can be retained by appropriate molecular design.

Results and discussion

Synthetic strategies

For systematic structure–activity relationship studies a large number of derivatives of jasmonic acid was required. Most of the compounds were synthesized starting from commercially available methyl jasmonate **1a** or jasmonic acid **1**. Only two derivatives of jasmonic acid (**33** and **43**) were prepared by more complicated synthetic routes. In order to refine our synthetic targets, we were interested in testing our synthesis products as soon as possible and, therefore, some reactions were not optimized. All compounds were obtained as mixtures of the C-2/C-3-*cis*/*trans*-isomers. The thermodynamically more stable *trans*-isomer is predominating as in jasmonic acid.[4] Synthetic targets were derived from structures of jasmonates previously isolated from plant material.

Naturally occurring jasmonates often contain oxidized pentenyl side chains or amino acid residues. Since it is assumed that these modifications are required for the specific biological activities of those molecules, we intended to synthesize compounds with varying side chains, possibly acting as mimics without being degraded in the regular way and, therefore, being metabolically more stable than their natural counterparts.

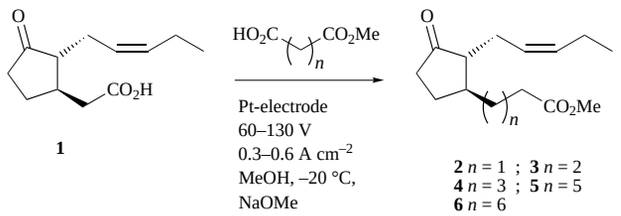
We decided to synthesize 3-oxaacyl compounds carrying ether functions instead of the nitrogen of the amino acid conjugates. Encouraged by the biological tests we prepared compounds with varying chain lengths for further investigation. In the light of the biological activity of long-chain jasmonates a homologous series of 3-acyljasmonates was prepared. Further, we decided to examine the influence of additional substituents in the C-3 side chain, since this moiety seems to have great influence on the test results.

In order to relate variations of the substituents to the biological activity we prepared jasmonates with variant C-2 side chains in a last series of syntheses. Since naturally occurring tuberonic acid glycoside has been described as an active compound, it seemed reasonable to investigate possible steric hindrance in the C-2 side chain. We chose to examine this by preparing dimeric compounds.

Synthesis of 3-acyl-2-pent-2-enylcyclopentanones

A homologous series of jasmonates bearing modified acyl side chains were prepared using the Kolbe reaction (Scheme 1).[5] The electrolysis was performed with jasmonic acid **1** and commercially available carboxy diacid mono esters using methanol as solvent. For deprotonation NaOMe was added. Reactions were completed when the reaction mixture became alkaline. This work was contemporaneous with that of Boland and Steckhan.[6]

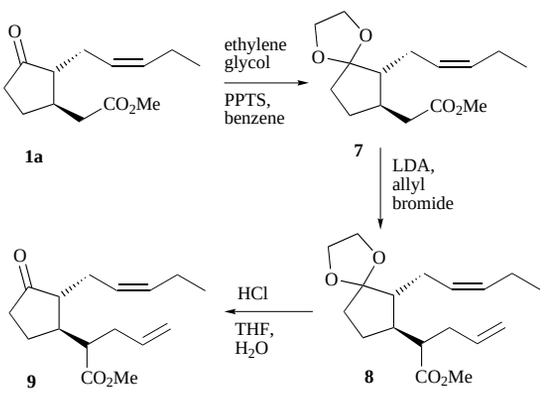
Using the Kolbe reaction two groups of derivatives of methyl jasmonates were prepared: (*a*) compounds **3** and **6** with a side chain bearing an even number of carbon atoms and (*b*) compounds **2**, **4** and **5** with an odd number of carbon atoms. In the course of jasmonate biosynthesis, only group (*a*) can be converted into methyl jasmonate by β-oxidation.



Scheme 1

Synthesis of analogues of methyl jasmonate with modified C-3 side chain

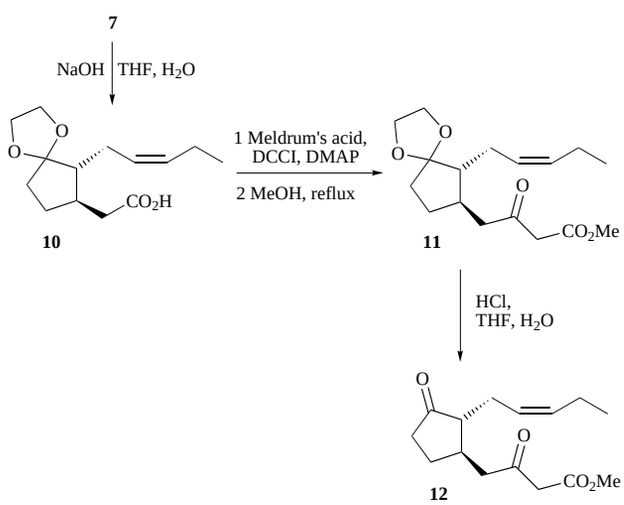
In order to introduce an allyl group in the acyl side chain methyl jasmonate **1a** needed to be protected as an acetal (Scheme 2).



Scheme 2

The keto group was converted quantitatively into the acetal with ethylene glycol and pyridinium tosylate in refluxing benzene.[7] The resulting compound **7** was allylated in the presence of lithium diisopropylamide and allyl bromide. Finally, the acetal was cleaved quantitatively under acidic conditions to give the derivative **9** which represents a jasmonate with an allylated C-3 side chain.

Another modification of the acyl side chain is its conversion into a β-keto ester. The synthesis started by saponifying protected methyl jasmonate **7** under basic conditions (Scheme 3).



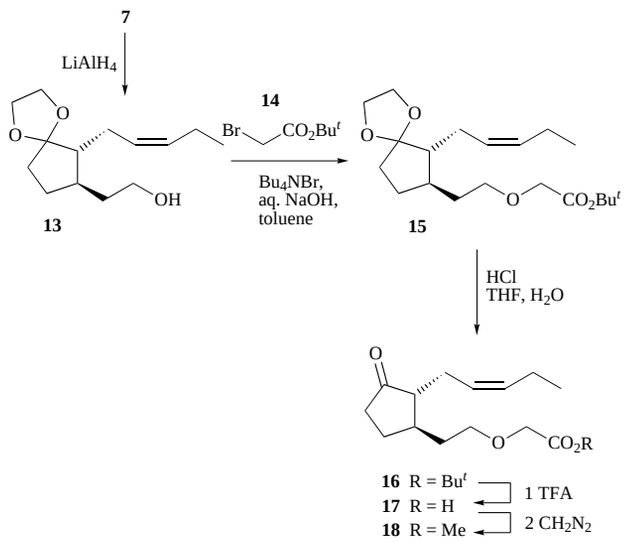
Scheme 3

In the presence of DCCI and DMAP, the resulting acid **10** was coupled with Meldrum's acid, 2,2-dimethyl-1,3-dioxane-4,6-dione.[9] The crude product was dissolved in dry methanol and refluxed for 1 h. Separation of the reaction mixture by flash chromatography afforded the β-keto ester **11** (42%).[9] In this case, Meldrum's acid can be regarded as a synthetic equivalent for deprotonated methyl acetate. Finally, the acetal was cleaved under acidic conditions to give the β-keto ester **12** which represents a jasmonate derivative with a β-keto acyl side chain.

Synthesis of 3-oxaacyl analogues of methyl jasmonate

Regarding further structure–activity studies, we were also interested in derivatives that are similar to the biosynthetic precursors of methyl jasmonate but cannot be converted into it. One possibility is the insertion of oxygen in the C-3 side chain thereby preventing β-oxidation that usually leads to methyl jasmonate.

The synthesis of the first 3-oxaacyl analogue started with the reduction of protected methyl jasmonate **7** with $LiAlH_4$ (Scheme 4).



Scheme 4

The resulting alcohol **13** was then coupled with *tert*-butyl bromoacetate **14** in a two-phase system consisting of aqueous NaOH and toluene in the presence of tetrabutylammonium bromide as phase transfer catalyst to give **15** (89%),[10] the acetal and the *tert*-butyl ester of which were subsequently cleaved under acidic conditions. Finally, the carboxy acid **17** was converted into the methyl ester **18** using diazomethane. Thus, although it can be seen that compound **18** is structurally similar to the pentanoyl derivative **4**, the presence of a 3-oxa group prevents β-oxidation.

Our next target was a compound with an elongated side chain at C-3. The conversion of alcohol **13** into the bromide **19** using tetrabromomethane and triphenylphosphine was the first step (Scheme 5).[11]

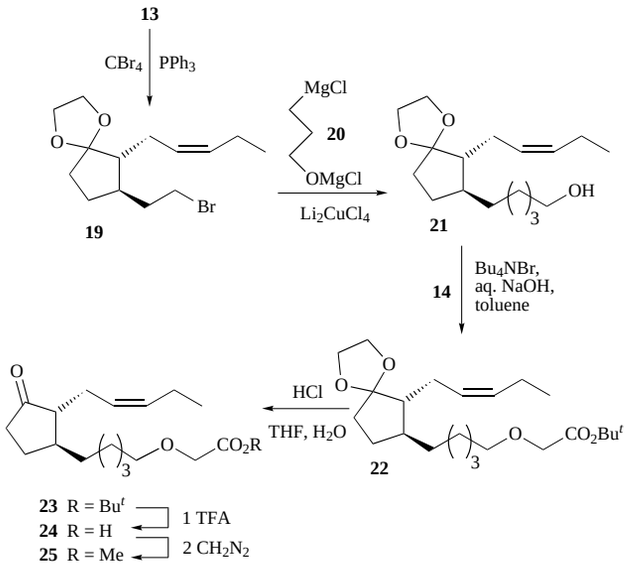
Next, the Grignard reagent **20**[12] was added to the bromide **19** to give **21** (76%).[13] After coupling of **21** with *tert*-butyl bromoacetate **14**, the resulting compound **22** was readily converted into the corresponding carboxy acid **24** and the methyl ester **25**, using similar methodology to that described above. It is apparent that compound **25** can be regarded as a jasmonate derivative with an octanoyl side chain whose biosynthetic degradation is prevented by the oxygen β to the carboxy group.

Our final 3-oxaacyl target was an analogue of methyl jasmonate with a side chain containing two oxygens (Scheme 6). The initial step comprised reducing the *tert*-butyl ester **15** with $LiAlH_4$.

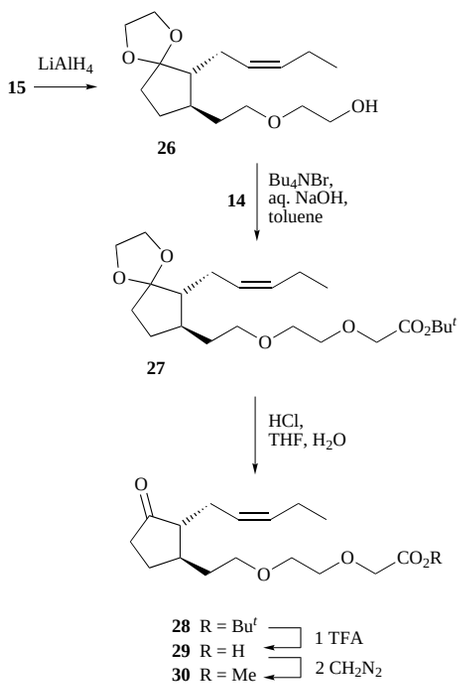
In a similar manner to that described above, the alcohol **26** was coupled with *tert*-butyl bromoacetate **14** to give **27** (79%). After cleavage of the acetal and the *tert*-butyl group, the carboxy acid **29** was converted into the methyl ester **30** using diazomethane.

Synthesis of analogues of methyl jasmonate with variant side chains at C-2

In order to analyze the influence of the olefinic side chain on

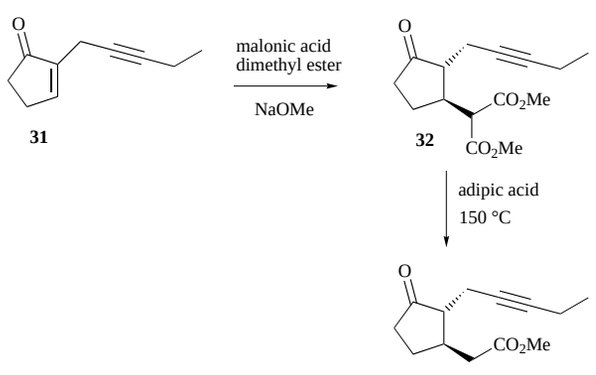


Scheme 5



Scheme 6

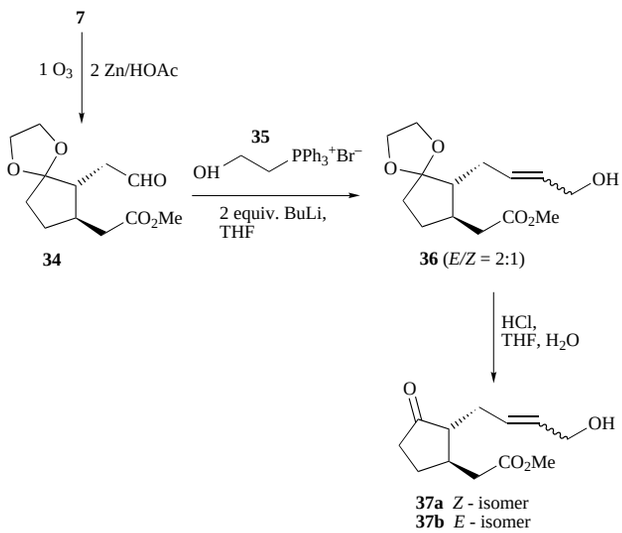
the biological activity of jasmonates, derivatives with modified side chains at C-2 were prepared. Our first target was a derivative of methyl jasmonate in which the pentenyl side chain was replaced by a pentynyl side chain (Scheme 7).



Scheme 7

The synthesis started with the substituted cyclopentenone **31** which was prepared according to the procedure of Tsuji *et al.*[14] Michael addition of dimethyl malonate to the cyclopentenone **31** gave **32** in good yield, which after decarboxylation gave the desired derivative **33**, a methyl jasmonate derivative with a pentynyl side chain.

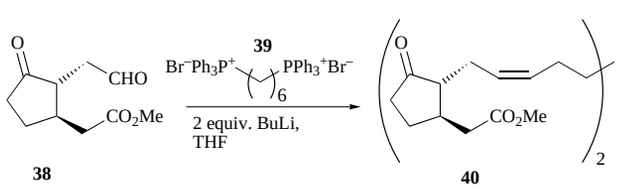
Biological studies revealed that jasmonic acid is metabolized by allylic oxidation of the C-2 side chain.[15] Thus, compounds **37a** and **37b** bearing a butenyl side chain with a hydroxy group were of particular interest (Scheme 8).



Scheme 8

To prepare these compounds, protected methyl jasmonate **7** was first ozonized in CH_2Cl_2 at −78 °C which, after reductive work-up with Zn–HOAc, gave the aldehyde **34** (81%).[16] Modification of the side chain was completed by a Wittig reaction according to the following methodology. A suspension of 2-hydroxyethyl(triphenyl)phosphonium bromide **35** in THF was treated with butyllithium (2 equiv.) in hexane. The aldehyde **34** was added at −30 °C to the reaction mixture and after work-up **36** was isolated (17%) as a 2:1 mixture of *E*/*Z* isomers.[17] The *E*/*Z* ratio was determined by NMR spectroscopy and GC. Finally, the acetal was cleaved under acidic conditions to afford **37** as a mixture of *E*/*Z*-isomers, which was separated by chromatography on Sephadex® to give pure *Z*- and *E*-isomer **37a** and **37b**, respectively.

A further interesting target was the dimeric jasmonate derivative **40** in which the pentenyl side chain is substituted with a second molecule of methyl jasmonate (Scheme 9).



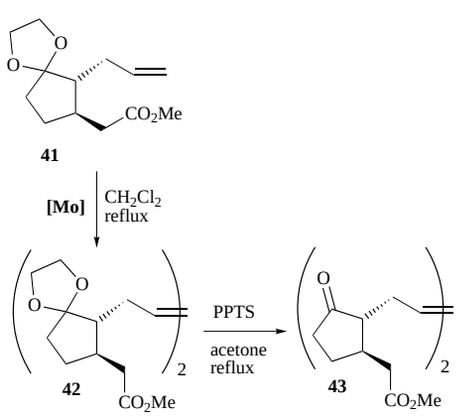
Scheme 9

Conveniently, Wittig reaction of the aldehyde **38**[16] with the phosphonium salt **39**, the latter deprotonated with butyllithium (2 equiv.), provided access to compound **40**.

Compound **43**, a homologue of **40**, was synthesized by a different strategy, namely by olefin metathesis (Scheme 10). In the presence of Schrock's molybdenum catalyst[18] [$PhMe_2$-CCH=Mo=N(2,6-$Pr^i_2C_6H_3$)][$OCMe(CF_3)_2$]$_2$ ([**Mo**]), the protected jasmonate derivative **41** having an allylic side chain,[19] dimerized in refluxing methylene dichloride under an atmosphere of Ar. Compound **42** was isolated (89%) as a 2:1 mixture of *E*/*Z* isomers. Deprotection of **42** in refluxing acetone in

Table 1 Biological effects of test compounds

Compound	*Eschscholzia* elicitation (Score)	*Bryonia* coiling (Score)	Senescence assays: Barley (% Chlorophyll loss)	Senescence assays: Tomato (% Chlorophyll loss)	Tomato transpiration [Relative rate (%)]
Homologous series of 3-acyl derivatives of 2-pent-2-enylcyclopentanone					
1a	++++	+++	42	28	77
2	+++	0	20	17	95
3	++++	++	30	25	67
4	++++	0	21	20	91
5	++++	++	18	9	87
6	+++	++++	37	19	88
Analogues of methyl jasmonate with modified C-3 side chain					
9	0	+++	27	35	98
12	++++	+	14	5	109
3-Oxaacyl analogues of methyl jasmonate					
18	++++	0	22	4	98
24	++	0	26	6	48
25	++	0	17	9	52
29	toxic	0	33	3	53
30	toxic	0	28	39	47
Analogues of methyl jasmonate with variant side chains at C-2					
33	++	0	28	9	64
37a	++	0	23	27	61
37b	++	0	32	25	90
40	++	0	28	9	72
43	0	0	26	1	92



Scheme 10

the presence of pyridinium tosylate gave **43** in good yield. Compound **43** can be regarded as a jasmonate in which the C-2 side chain is replaced by a shortened molecule of methyl jasmonate. Olefin cross-metathesis, a new catalytic method for carbon–carbon double bond formation, has previously been used in our laboratories to synthesize different derivatives of jasmonic acid.[19]

Biological activity

The spectrum of biological responses to jasmonic acid/methyl jasmonate is wide and covers, in addition to the desired effects of inducing plant defence responses, unacceptable side effects such as reduction of transpiration (an adverse effect on a plant's stomatal pores), a strong promotion of senescence and, as well, an inhibition of plant growth.[1] Nothing is known on the structural requirements for these individual jasmonate responses and, in attempts to develop lead structures appropriate as plant defence regulators for practical applications, it is of the utmost importance to eliminate, as much as possible, any unwanted physiological side effects of those substances, while not negatively affecting the induction of the defence response. Thus, in addition to synthesizing a wide array of jasmonate derivatives, it was necessary to test these derivatives on a carefully chosen set of diverse bioassays, covering all those known undesirable side effects of natural jasmonates.

The tendril coiling assay[20] rapidly identifies compounds with growth inhibitory potential, while the detached leaf segment (barley) and whole plant (tomato) loss-of-chlorophyll assays would reveal senescence promoters whereas the tomato transpiration assay focusses on compounds that may adversely affect plant–gas exchange. The *Eschscholzia californica* bioassay is a well established and sensitive assay for the induction of low-molecular-weight defence compounds. It is, in fact, the system with which the induction of phytoalexins by methyl jasmonate was first discovered.[3,21,22,23] Phytoalexins are active in protecting plants against pathogens and herbivores,[24] and their induction, therefore, serves as an indication of plant protection induced by the various jasmonate analogues tested herein.

It can be seen from the results (Table 1) that structure–activity relationships are different for each of the bioassay systems compared, and in some cases drastically so. While most of the compounds showed some activity in the *E. californica* elicitation assay as well as in the barley and tomato assays, a much smaller group of test compounds was active in the tendril coiling assay. In this latter assay, a particularly interesting structure–activity relationship exists within the group of 3-acyl homologues of methyl jasmonate **1a**. Compared to **1a**, the propionyl analogue **2** was found to be completely inactive, stressing the importance of an acetyl side chain for biological activity. The butyryl homologue **3** was again active, but weaker than methyl jasmonate **1a**, whilst the pentanoyl derivative **4** was again inactive. Thus, the activity of **3** in all probability must be traced to its β-oxidation to jasmonic acid. Interestingly the heptanoyl analogue **5** was active. β-Oxidation of **5** would yield the C_5 and then the C_3 analogues of jasmonic acid **1**, both of which (applied as the methyl esters **2** and **4**) were inactive. Thus the activity of **5** must be genuine and cannot stem from its conversion into an active metabolite. In line with this, is the finding that the octanoyl homologue **6** is strongly active, in fact significantly more active than methyl jasmonate **1a** itself.[3,21] This high activity makes it unlikely that the activity of **6** stems

from its conversion by β-oxidation which would yield jasmonic acid. These findings are in agreement with the activity data reported earlier for 12-oxophytodienoic acid[3,21] an 11,12-dehydroanalogue of **6**, and that of coronatine[21] a structural analogue of 12-oxophytodienoic acid. Thus, the data for the tendril coiling assay clearly define two structurally non-overlapping groups of active compounds, one centred around the structure of methyl jasmonate **1a** and the second centred around the structure of **6**, the free acid of which occurs as an intermediate in the biosynthesis of jasmonic acid and which is structurally closely similar to that of 12-oxophytodienoic acid and coronatine.

In contrast to this, all compounds within the 3-acyl homologue series are highly active in the *E. californica* elicitation assay. In this assay, the length of the side chain attached to C-3 can thus not be a prime determinant of biological activity. There are, however, some limitations to the C-3 side chain, as demonstrated by the fact that **9** is inactive in this assay while it was as active as methyl jasmonate **1a** in the tendril coiling assay.

It is thus possible to separate structurally the elicitation properties from the growth inhibitory potential of a jasmonate by appropriate molecular design. For example, **9** is strongly active in the coiling assay but does not elicit the cell culture. In contrast, **4** is a clear elicitor, although completely inactive in the coiling assay.

Oxa analogues appear to behave similarly, if not identically, with the parent compounds, as exemplified for **18**, **24** and **25** which are active in the cell culture elicitation, but inactive in the tendril coiling assay. The activity of **18**, **24** and **25** on the cell culture are evidence that, in this assay also, β-oxidation of the 3-acyl side chain to an acetyl moiety is not prerequisite to biological activity.[22]

Being completely inactive in the coiling assay, analogues with variant side chains at C-2 such as **7**, **37a**, **37b** and **40** show a clear reaction in cell culture elicitation test which is, however, significantly weaker than that of methyl jasmonate **1a**. Apparently, modification of the olefinic side chain of jasmonates can be used to design analogues with a specific biological activity as well.

Inspection of the data in Table 1 further shows that the senescence-promoting activity of a compound varies independently of its activity in the other assays and even among the two senescence assays used here. As examples, **18** and **12** do respond as strongly as methyl jasmonate **1a** in the cell culture elicitation assay but have, in contrast to **1a**, almost no senescence promoting activity. Conversely, **9** stimulates senescence to almost the same extent as **1a**, but is completely inactive on the cell culture. The data in Table 1 further show that the ability to alter rates of transpiration does not correlate with any of the aforementioned biological activities. As a general conclusion, it can be stated that, while the molecular basis for the various biological activities probed in this study remain unknown, molecular design of a jasmonate analogue will allow tailoring of its biological activity profile in order to reduce or eliminate unwanted side effects. An example for such a compound is **18**.

Experimental

General

MS spectra and high resolution mass spectra: Finnigan MAT 711 and MAT 955Q mass spectrometer (EI) with an ionization potential of 70 eV. Infrared spectra: Nicolet 750 FT infrared spectrometer. ^{1}H NMR spectra: Bruker AM 400, AM 270 and AC 200; chemical shifts relative to $CDCl_3$. ^{13}C NMR spectra including DEPT: Bruker AM 400, AM 270 and AC 200. Thin-layer chromatography was performed on Merck $^{60}F_{240}$ (0.2 mm) sheets which were then visualized with a solution of molybdophosphoric acid in acetic acid, under UV light or with an aqueous solution of $KMnO_4$. Preparative flash chromatography was performed on Merck (0.04–0.063 mm) silica gel using a positive pressure of air. PE = light petroleum (bp 40–60 °C) and MTBE = methyl *tert*-butyl ether. GC: Hewlett Packard 5890 Series II. Unless otherwise noted, all chemicals were of the highest commercially available purity and were used without further purification.

Eschscholzia californica elicitation

Cell suspension cultures of *E. californica* were provided by the cell culture laboratory of the Lehrstuhl für Pharmazeutische Biologie, Universität München. Cultures were routinely grown in 1 l conical flasks containing Linsmaier–Skoog medium (400 cm^3)[25] over 7 days at 23 °C on a gyratory shaker (100 rpm) in diffuse light (750 lux). For elicitation experiments, 24 g fresh weight (600 mg dry weight) of cells were inoculated into 150 cm^3 fresh Linsmaier–Skoog medium in which these cells were allowed to grow for 3 days under the above conditions. To each well of a 24-well multi-dish (Nunc), 1 cm^3 of the 3-day-old cell culture was aseptically pipetted. Elicitation of *E. californica* cell suspension cultures was achieved by the aseptic addition of methyl jasmonate or synthetic analogue diluted in 80% ethanol to a final concentration of 0.1–300 μM to the medium of each well. Each substance was tested in triplicate. Control wells were given an equivalent volume of 80% ethanol. To each was then added sterile water (250 μl) to prevent desiccation. The cell cultures were then incubated an additional 5 days on a reciprocal shaker at 200 strokes min^{-1} at 23 °C under diffuse light (750 lux).

After 5 days, the cell suspension in each well of the 24-well multi-dish was transferred to a 1,5 cm^3 microcentrifuge tube and the cells were collected by centrifugation at 14 000 g for 5 min at room temperature. The supernatant was discarded and the cell pellet was immediately extracted with 80% ethanol (1 cm^3) containing 0.5% HCl for 2 h at 60 °C. The cell debris was removed by centrifugation at 14 000 g for 5 min at room temperature and the supernatant (250 μl) was placed into one well of a 96-well microtiter plate (Nunc). The extinction at 490 nm of each extract was determined with a Microplate Reader (MR600, Dynatech) and was converted into total benzophenanthridine alkaloids per cm^3 of cell culture using a calibration curve determined with extinction measurements using authentic alkaloid standards.[22] Scores: ++++, 5–10-fold more alkaloid accumulated than in control; +++, 3–5-fold more alkaloid accumulated than in control; ++, 2–3-fold more alkaloid accumulated than in control; +, 1.5–2-fold more alkaloid accumulated than in control; 0, equivalent quantity of alkaloid accumulated as in control.

Tendril coiling bioassay

This assay was performed exactly as described previously.[20] For each test compound, the analysis was repeated three times on different days. For each assay, per data point, two duplicates of three detached tendrils were used. Scoring was done independently for each assay and the results presented are averaged scores of the three assays. Scores: ++++, reaction stronger than that of methyl jasmonate; +++ reaction comparable to that of methyl jasmonate; ++, a clear reaction, but significantly weaker than that of methyl jasmonate; +, a marginal reaction clearly above background; 0, no reaction above background activity.

Tomato bioassays

Lycopersicon esculentum Mill. cv. First-in-the-Field was raised in a greenhouse until 4 weeks old. Plants were then transferred into a phytotron chamber (photoperiod 8 h, T_{day} 20 °C, T_{night} 17 °C, 70% relative humidity, 70–90 μmol photons m^{-2} s^{-1} photosynthetically active radiation) and grown until 6 weeks old (15 cm height). Test compounds (10 μM) dissolved in 40% acetone (v/v) containing 0.1% (w/v) Tween 20, were then sprayed on the leaves until run-off. Three plants were sprayed per data point, and the experiment was repeated twice on

separate occasions. After spraying, plants were incubated for 24 h and then watered with an excess of water that was allowed to drip off for 1 h. The pots were then enclosed in 31 polythene bags taped tightly around the stems. Plants were then weighed every 2 h for 8 h to determine the rates of transpiration. The procedure was repeated the next day starting with excess watering of the plants, covering of the plants with the polythene bag and weight determinations (only the second set of transpiration data is shown in the Results section, and all data were normalized for leaf mass and calculated initially as cm^3 of water lost per gram of leaf fresh weight). Data are given as relative rates of transpiration compared to control plants treated with spray solution only, and then processed identically with all other plants.

After collection of the transpiration data, plants were further incubated until 72 h post application of the test compounds. Leaves were then harvested, weighed and extracted for chlorophyll determination according to Arnon.[26] Senescence data are given as % chlorophyll lost compared to control plants treated with spray solution only.

Barley bioassay

Hordeum vulgare L. cv. Baronesse was grown in 20 × 40 cm trays in standard soil in a phytotron chamber (climatic parameters identical with those of the tomato, see above) until 10 days old. Primary-leaf segments were obtained by excising a 3-cm leaf segment starting 2 cm below the tip. Per data point, 16 leaf segments were co-incubated in a 9 cm diameter Petri dish containing 50 μM kinetin (10 cm^3) and the appropriate test compound (10 μM), dissolved in methanol [final concentration <0.1% (v/v), controls: solvent only]. The leaf segments were then incubated at 25 °C in the dark for 40 h, after which they were extracted and chlorophyll determined according to Arnon.[26] Data from two independent experiments were averaged and are expressed as relative loss of chlorophyll as percent of controls.

Methyl 3-(3-oxo-2-pent-2-enylcyclopentyl)propionate 2: general procedure for the Kolbe reaction

Jasmonic acid **1** (200 mg, 0.95 mmol) and monomethyl malonate (560 mg, 4.7 mmol) were dissolved in methanol (2 cm^3) to which sodium (10 mg, 0.43 mmol) was then added. The mixture was cooled to −20 °C and electrolyzed with platinum electrodes at 0.3 A cm^{-2} until alkaline reaction was observed. The resulting mixture was poured into water (8 cm^3) and extracted with MTBE. The combined extracts were dried ($MgSO_4$) and evaporated *in vacuo* and dimethyl succinate was distilled off in a Kugelrohr apparatus. Flash chromatography (MTBE–PE, 1 : 3) of the residue on silica gel afforded **2** (30 mg, 0.12 mmol, 13%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1734 (ester and ketone); δ_H(270 MHz, $CDCl_3$) 0.95 (3H, t, *J* 7.5), 1.38 (1H, m), 1.56 (1H, m), 1.8–1.9 (2H, m), 2.0–2.2 (5H, m), 2.37 (4H, m), 3.68 (3H, s), 5.24 (1H, dtt, *J* 11, 7 and 1.5) and 5.44 (1H, dtt, *J* 11, 7 and 1.5); δ_C(67.5 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 25.2 (CH_2), 26.6 (CH_2), 29.6 (CH_2), 31.7 (CH_2), 37.8 (CH_2), 40.5 (CH), 51.5 (CH), 54.7 (CH_3), 125.0 (CH), 133.7 (CH), 173.7 (C_q) and 220.0 (C_q); *m*/*z* 238.1563 (M^+, 38%. $C_{14}H_{22}O_3$ requires 238.1569), 220 (18%), 207 (22), 191 (20), 151 (74), 109 (40), 96 (60) and 83 (100).

Methyl 4-(3-oxo-2-pent-2-enylcyclopentyl)butyrate 3

Following the general procedure described above for compound **2**, jasmonic acid **1** (330 mg, 1.57 mmol) and monomethyl succinate (900 mg, 6.8 mmol) were electrolyzed in methanol (3 cm^3) and sodium methanolate (20 mg, 0.37 mmol) at 0.6 A cm^{-2}. Dimethyl adipate was distilled off in a Kugelrohr apparatus, and the residue flash chromatographed (MTBE–PE, 1 : 3) to afford **3** (55 mg, 0.22 mmol, 14%) as a colourless oil; ν_{max}(ATR)/cm^{-1} 2959, 1738 (ester and ketone) and 1164; δ_H(270 MHz, $CDCl_3$) 0.98 (3H, t, *J* 7.5), 1.2–1.5 (3H, m), 1.52–1.92 (6H, m), 1.98–2.24 (4H, m), 2.26–2.42 (6H, m), 3.66 (3H, s), 5.24 (1H, dtt, *J* 11, 7 and 1.5) and 5.44 (1H, dtt, *J* 11, 7 and 1.5); δ_C(67.5 MHz, $CDCl_3$) 14.0 (CH_3), 20.4 (CH_2), 22.4 (CH_2), 25.2 (CH_2), 26.8 (CH_2), 29.6 (CH_2), 34.0 (CH_2), 37.8 (CH_2), 40.7 (CH), 51.4 (CH), 54.7 (CH_3), 125.1 (CH), 123.5 (CH), 173.8 (C_q) and 220.2 (C_q); *m*/*z* 252.1725 (M^+, 30%. $C_{15}H_{24}O_3$ requires 252.1725), 234 (10%), 184 (10), 151 (80), 133 (15), 109 (20), 95 (30) and 83 (100).

Methyl 5-(3-oxo-2-pent-2-enylcyclopentyl)pentanoate 4

Following the general procedure described above for compound **2**, jasmonic acid **1** (250 mg, 1.2 mmol) and monomethyl glutarate (700 mg, 4.8 mmol) were electrolyzed in methanol (3 cm^3) and sodium methanolate (20 mg, 0.37 mmol) at 0.6 A cm^{-2}. Dimethyl octanoate was distilled off in a Kugelrohr apparatus, and the residue flash chromatographed (MTBE–PE, 1 : 6) to afford **4** (55 mg, 0.2 mmol, 17%) as a colourless oil; ν_{max}(ATR)/cm^{-1} 2933, 1738 (ester and ketone) and 1163; δ_H(270 MHz, $CDCl_3$) 0.96 (3H, t, *J* 7.5), 1.2–1.5 (6H, m), 1.56–1.92 (5H, m), 2–2.2 (3H, m), 2.28–2.4 (4H, m), 3.66 (3H, s), 5.24 (1H, dtt, *J* 11, 7 and 1.5) and 5.44 (1H, dtt, *J* 11, 7 and 1.5); δ_C(67.5 MHz, $CDCl_3$) 14.0 (CH_3), 20.4 (CH_2), 24.9 (CH_2), 25.3 (CH_2), 26.5 (CH_2), 26.9 (CH_2), 33.8 (CH_2), 34.2 (CH_2), 37.9 (CH_2), 40.8 (CH), 51.4 (CH), 54.8 (CH_3), 125.3 (CH), 133.4 (CH), 173.9 (C_q) and 220.4 (C_q); *m*/*z* 266.1888 (M^+, 15%. $C_{16}H_{26}O_3$ requires 266.1881), 248 (10%), 151 (55), 133 (15), 109 (20), 95 (35) and 83 (100).

Methyl 7-(3-oxo-2-pent-2-enylcyclopentyl)heptanoate 5

Following the general procedure described above for compound **2**, jasmonic acid **1** (650 mg, 3.1 mmol) and methyl heptanoate (860 mg, 4.9 mmol) were electrolyzed in methanol (4 cm^3) and sodium methanolate (30 mg, 0.55 mmol) at 0.6 A cm^{-2}. Dimethyl dodecanoate was distilled off in a Kugelrohr apparatus, and the residue flash chromatographed (MTBE–PE, 1 : 6) to afford **5** (260 mg, 0.88 mmol, 28%) as a colourless oil; ν_{max}(ATR)/cm^{-1} 1739 (ester and ketone), 1168 and 2929; δ_H(270 MHz, $CDCl_3$) 0.96 (3H, t, *J* 7.5), 1.2–1.5 (8H, m), 1.58–1.75 (4H, m), 1.8 (1H, m), 1.98–2.2 (4H, m), 2.3–2.4 (5H, m), 3.68 (3H, s), 5.24 (1H, dtt, *J* 11, 7 and 1.5) and 5.44 (1H, dtt, *J* 11, 7 and 1.5); δ_C(100 MHz, $CDCl_3$) 14.0 (CH_3), 20.4 (CH_2), 24.7 (CH_2), 25.2 (CH_2), 26.7 (CH_2), 26.9 (CH_2), 28.9 (CH_2), 29.3 (CH_2), 33.9 (CH_2), 34.5 (CH_2), 37.8 (CH_2), 40.9 (CH), 51.3 (CH), 54.8 (CH_3), 125.3 (CH), 133.5 (CH), 174.0 (C_q) and 220.4 (C_q); *m*/*z* 294.2195 (M^+, 10%. $C_{18}H_{30}O_3$ requires 294.2195), 276 (10%), 263 (10), 226 (26), 151 (50), 124 (35), 95 (35) and 83 (100).

Methyl 8-(3-oxo-2-pent-2-enylcyclopentyl)octanoate 6

Following the general procedure described above for compound **2**, jasmonic acid **1** (270 mg, 1.3 mmol) and methyl octanoate (550 mg, 2.9 mmol) were electrolyzed in methanol (3 cm^3) and sodium methanolate (20 mg, 0.37 mmol) at 0.6 A cm^{-2}. Dimethyl tetradecanoate was partially distilled off in a Kugelrohr apparatus, and the residue flash chromatographed (MTBE–PE, 1 : 10) to afford **6** (40 mg, 0.12 mmol, 10%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3029, 2964, 1735 (ester and ketone), 1231 and 1098; δ_H(400 MHz, $CDCl_3$) 0.96 (3H, t, *J* 7.5), 1.2–1.45 (10H, m), 1.6–1.71 (3H, m), 1.75–1.9 (2H, m), 2.0–2.2 (4H, m), 2.34 (5H, m), 3.66 (3H, s), 5.24 (1H, dtt, *J* 11, 7 and 1.5) and 5.42 (1H, dtt, *J* 11, 7 and 1.5); δ_C(67.5 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 24.8 (CH_2), 25.3 (CH_2), 26.9 (CH_2), 27.0 (CH_2), 29.0 (CH_2), 29.1 (CH_2), 29.5 (CH_2), 33.9 (CH_2), 34.6 (CH_2), 37.9 (CH_2), 41.0 (CH), 51.4 (CH), 54.9 (CH_3), 125.3 (CH), 133.3 (CH), 174.2 (C_q) and 220.7 (C_q); *m*/*z* 308 (M^+, 8%), 290 (6), 240 (22), 151 (53), 124 (44), 95 (36) and 83 (100).

Methyl (6-pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)acetate 7

To a solution of **1a** (5 g, 22.3 mmol) in benzene (50 cm^3), were added ethylene glycol (10 g, 161.3 mol) and pyridinium tosylate (1 g, 4 mmol) and the mixture was refluxed for 16 h, with water

separation by means of a Dean–Stark trap. Excess of solvent was then removed *in vacuo* from the mixture which was diluted with MTBE (100 cm^3), washed with sat. aqueous $NaHCO_3$, water and brine, dried ($MgSO_4$) and evaporated under reduced pressure to give **7** (5.87 g, 21.9 mmol, 98%); $\nu_{max}(CHCl_3)/cm^{-1}$ 1732 (ketone) and 948; δ_H(400 MHz, $CDCl_3$) 0.96 (3H, t, *J* 7.5), 1.3 (1H, qd, *J* 12.5 and 8), 1.64–1.8 (3H, m), 1.92 (1H, ddt, *J* 8.5, 8 and 5), 2.0–2.15 (4H, m), 2.24 (1H, m), 2.31 (1H, dd, *J* 15 and 10), 2.59 (1H, dd, *J* 15 and 4.5), 3.65 (3H, s), 3.8–3.95 (4H, m) and 5.36 (2H, m); δ_C(50 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 26.4 (CH_2), 27.9 (CH_2), 35.1 (CH_2), 39.3 (CH), 39.9 (CH_2), 49.1 (CH), 51.1 (CH_3), 64.1 (CH_2), 64.6 (CH_2), 117.6 (C_q), 123.6 (CH), 132.0 (CH) and 173.1 (C_q); *m/z* 268.1675 (M^+, 20%. $C_{15}H_{24}O_4$ requires 268.1668), 237 (9%), 195 (18), 153 (20) and 99 (100).

Methyl 2-(6-pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)pent-4-enoate 8

To a solution of diisopropylamine (60 mg, 0.6 mmol) in dry THF (10 cm^3) was added butyllithium (1.6 M in hexane; 350 µl) at 0 °C. After being stirred for 5 min, the mixture was cooled to −78 °C and **7** (150 mg, 0.56 mmol) was added to it. The mixture was stirred for 30 min and after which 1-bromoprop-2-ene (67 mg, 0.56 mmol) was added to it. The solution was allowed to warm to room temperature during 3 h, after which it was poured into water (10 cm^3), acidified with 1 M aqueous HCl (1 cm^3) and extracted with MTBE. The combined extracts were dried ($MgSO_4$) and evaporated *in vacuo* and the residue was flash chromatographed on silica gel (MTBE–PE, 1:5) to afford **8** (129 mg, 0.42 mmol, 75% yield) as a colourless oil; ν_{max}(ATR)/cm^{-1} 2960, 1735 (ester) and 1163; δ_H(270 MHz, $CDCl_3$) 0.96 (1H, t, *J* 7), 1.4 (1H, m), 1.85–1.9 (4H, m), 1.95–2.4 (4H, m), 2.55 (1H, ddd, *J* 4, 7 and 9), 3.86 (3H, s), 3.9 (4H, m), 5.02 (2H, m), 5.35 (2H, m) and 5.7 (1H, m); δ_C(67.5 MHz, $CDCl_3$) 14.0 (CH_3), 20.3 (CH_2), 24.8 (CH_2), 27.5 (CH_2), 32.7 (CH_2), 34.5 (CH_2), 44.1 (CH), 48.6 (CH), 49.4 (CH), 51.0 (CH_3), 63.6 (CH_2), 64.5 (CH_2), 116.2 (CH_2), 117.4 (C_q), 127.3 (CH), 131.9 (CH), 135.6 (CH) and 174.9 (C_q); *m/z* 308.1988 (M^+, 8%. $C_{18}H_{28}O_4$ requires 308.1987), 277 (10%), 239 (10), 195 (95), 153 (30), 99 (100) and 86 (90).

Methyl 2-(3-oxo-1-pent-2-enylcyclopentyl)pent-4-enoate 9: general procedure for the cleavage of acetals

A solution of **8** (100 mg, 0.325 mmol) , THF (3 cm^3) and 2 M HCl (2 cm^3) was stirred at room temperature for 3 h after which it was treated with a solution of aq. $NaHCO_3$ (5 cm^3) and extracted with MTBE. The extract was dried ($MgSO_4$) and evaporated *in vacuo*. The residue was chromatographed on silica gel (MTBE–PE, 1:2) to give **9** (77 mg, 0.29 mmol, 90%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 2963, 1738 (ester and ketone) and 1164; δ_H(400 MHz, $CDCl_3$) 0.96 (3H, t, *J* 8), 1.6 (1H, m), 2.0–2.5 (11H, m), 2.58 (1H, ddd, *J* 11, 7 and 4), 3.7 (3H, s), 5.0–5.15 (2H, m), 5.22 (1H, dtt, *J* 11, 7 and 1.5), 5.44 (1H, dtt, *J* 11, 7 and 1.5) and 5.66 (1H, tdd, *J* 10, 7 and 3.5); δ_C(67.5 MHz, $CDCl_3$) 13.9 (CH_3), 20.7 (CH_2), 24.5 (CH_2), 25.6 (CH_2), 32.7 (CH_2), 37.5 (CH_2), 41.8 (CH), 48.6 (CH), 49.4 (CH), 51.0 (CH), 51.8 (CH_3), 116.8 (CH_2), 124.5 (CH), 134.5 (CH), 135.5 (CH), 174.4 (C_q) and 218.8 (C_q); *m/z* 264.1725 (M^+, 3%. $C_{16}H_{24}O_3$ requires 264.1725), 233 (5%), 194 (10), 151 (100), 109 (15) and 83 (30).

(6-Pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)acetic acid 10

A solution of **7** (600 mg, 2.2 mmol), NaOH (200 mg, 5 mmol), water (15 cm^3) and THF (10 cm^3) was stirred at room temperature for 2 h after which it was acidified with 1 M aqueous HCl (pH 3) and extracted with MTBE. The extract was washed with brine, dried ($MgSO_4$) and evaporated under reduced pressure to give **10** (550 mg, 2.16 mmol, 98%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3187 (OH) and 1706 (carboxy acid); δ_H(400 MHz, $CDCl_3$) 0.96 (3H, t, *J* 7.5), 1.22–1.43 (2H, m), 1.61–1.81 (3H, m), 1.93–2.16 (4H, m), 2.17–2.33 (3H, m), 2.65 (1H, dd, *J* 16 and 4), 3.80–3.96 (4H, m) and 5.3–5.42 (2H, m); δ_C(50 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 26.5 (CH_2), 28.0 (CH_2), 35.1 (CH_2), 39.1 (CH), 39.9 (CH_2), 51.2 (CH), 64.1 (CH_2), 64.7 (CH_2), 117.7 (C_q), 127.5 (CH), 132.3 (CH) and 178.9 (C_q); *m/z* 254 (M^+, 10%), 225 (5), 195 (20), 153 (22), 99 (100) and 86 (20).

Methyl 3-oxo-4-(6-pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)-butyrate 11

A solution of **10** (1.035 g, 4.07 mmol), DCCI (902 mg, 4.38 mmol), DMAP (801 mg, 6.56 mmol) and Meldrum's acid (600 mg, 4.16 mmol) in dry CH_2Cl_2 (30 cm^3) was stirred at room temperature for 16 h. The mixture was filtered and evaporated *in vacuo* and the residue was dissolved in ethyl acetate (30 cm^3). The solution was washed with 1 M aqueous HCl, water and brine, dried ($MgSO_4$) and evaporated under reduced pressure. The residue was dissolved in dry methanol (15 cm^3) and the solution stirred at 65 °C for 1 h. The reaction mixture when flash chromatographed (MTBE–PE, 1:2) afforded **11** (532 mg, 1.73 mmol, 42%) as a colourless oil. As a by-product **7** (212 mg, 0.78 mmol, 20%) was isolated; $\nu_{max}(CHCl_3)/cm^{-1}$ 1751 (ester), 1721 and 1654 (β-keto ester) and 1235; δ_H(400 MHz, $CDCl_3$) 0.94 (3H, t, *J* 8), 1.13–1.25 (1H, m), 1.6–1.8 (3H, m), 1.91–2.27 (6H, m), 2.47 (1H, dd, *J* 17 and 10), 2.82 (1H, dd, *J* 17 and 4), 3.42 (2H, s), 3.73 (3H, s), 3.81–3.94 (4H, m) and 5.29–5.43 (2H, m); δ_C(50 MHz, $CDCl_3$) 14.1 (CH_3), 20.4 (CH_2), 26.4 (CH_2), 28.0 (CH_2), 35.0 (CH_2), 38.0 (CH), 49.0 (CH_2), 49.1 (CH_2), 51.1 (CH), 52.2 (CH_3), 64.1 (CH_2), 64.4 (CH_2), 117.4 (C_q), 127.6 (CH), 132.2 (CH), 167.4 (C_q) and 201.8 (C_q); *m/z* 310.1780 (M^+, 4%. $C_{17}H_{26}O_5$ requires 310.1780), 237 (4%), 195 (22), 194 (8), 153 (12), 125 (16), 99 (100), 86 (18), 67 (18) and 55 (18).

Methyl 3-oxo-4-(3-oxo-2-pent-2-enylcyclopentyl)butyrate 12

Following the general procedure described above for compound **9**, the acetal **11** (444 mg, 1.43 mmol) was cleaved in the presence of THF and aq. HCl. The reaction mixture when flash chromatographed (MTBE–PE, 1:2) afforded **12** (286 mg, 1.08 mmol, 76%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1745 (ester and ketone), 1723 and 1653 (β-keto ester) and 1237; δ_H(400 MHz, $CDCl_3$) 0.94 (3H, t, *J* 8), 1.38–1.45 (1H, m), 1.82–1.90 (1H, m), 1.97–2.18 (3H, m), 2.23–2.48 (5H, m), 2.58 (1H, dd, *J* 18 and 9), 2.94 (1H, dd, *J* 18 and 4), 3.45 (2H, s), 3.75 (3H, s), 5.25 (1H, dtt, *J* 11, 7 and 1) and 5.45 (1H, dtt, *J* 11, 7 and 1); δ_C(50 MHz, $CDCl_3$) 14.0 (CH_3), 20.5 (CH_2), 25.5 (CH_2), 27.2 (CH_2), 36.7 (CH), 37.6 (CH_2), 47.5 (CH_2), 49.2 (CH_2), 52.3 (CH_3), 53.7 (CH), 125.0 (CH), 134.0 (CH), 167.3 (C_q), 201.1 (C_q) and 218.7 (C_q); *m/z* 266.1518 (M^+, 2%. $C_{15}H_{22}O_4$ requires 266.1518), 208 (20%), 190 (12), 150 (100), 147 (40), 140 (32), 121 (72), 109 (42), 95 (46), 83 (90), 82 (96) and 67 (36).

2-(6-Pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)ethanol 13

A solution of **7** (2 g, 7.5 mmol) in THF (20 cm^3) was added to a suspension of $LiAlH_4$ (0.43 g, 11.3 mmol) in dry THF (30 cm^3). The mixture was stirred at room temperature for 2 h, after which it was cooled to 0 °C and treated with ice–water until hydrogen evolution stopped. The precipitate was filtered off and the filtrate was extracted with MTBE. The combined extracts were dried ($MgSO_4$) and evaporated *in vacuo*. Flash chromatography of the residue on silica gel (MTBE–PE 1:1) afforded **13** (1.49 g, 6.2 mmol, 82%) as a yellow oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3625, 3030 and 1039; δ_H(400 MHz, $CDCl_3$) 0.96 (3H, t, 7.5), 1.47 (1H, dddd, *J* 13, 9, 7 and 5.5), 1.6–1.91 (7H, m), 2.05 (2H, m), 2.07 (1H, dd, *J* 5.5 and 7.5), 2.24 (1H, m), 3.58–3.72 (2H, m), 3.82–3.93 (4H, m) and 5.38 (2H, m); δ_C(100 MHz, $CDCl_3$) 14.1 (CH_3), 20.4 (CH_2), 26.7 (CH_2), 27.9 (CH_2), 35.3 (CH_2), 38.6 (CH_2), 39.4 (CH), 51.7 (CH), 61.3 (CH_2), 63.9 (CH_2), 64.5 (CH_2), 117.9 (C_q), 128.1 (CH) and 131.8 (CH); *m/z* 240.1719 (M^+, 20%. $C_{14}H_{24}O_3$ requires 240.1719), 195 (21%), 153 (33), 114 (36) and 99 (100).

***tert*-Butyl 2-{2-(2-pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)-ethoxy}acetate 15: general procedure**

A mixture of the alcohol **13** (1 g, 4 mmol), toluene (30 cm^3), aq. NaOH (50%; 10 cm^3), tetrabutylammonium bromide (0.1 g, 0.4 mmol) and *tert*-butyl bromoacetate **14** (3.25 g, 16.7 mmol) was stirred at room temperature for 12 h. After neutralization with aq. HCl the resulting mixture was extracted with MTBE. The extract was dried ($MgSO_4$) and evaporated *in vacuo* and the residue was chromatographed on silica gel (MTBE–PE, 1:4) to give **15** (1.2 g, 3.4 mmol, 89%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1744 (ester) and 1135; δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, 7.5), 1.26 (1H, dq, *J* 8.5 and 12), 1.47 (9H, s), 1.6–2.1 (10H, m), 2.22 (1H, ddd, *J* 5.5, 6.5 and 14), 3.47–3.56 (2H, m), 3.8–3.85 (6H, m) and 5.35 (2H, m); δ_C(100 MHz, $CDCl_3$) 14.0 (CH_3), 20.4 (CH_2), 26.5 (CH_2), 27.9 (CH_2), 28.1 (CH_3), 35.2 (CH_2), 35.4 (CH_2), 39.8 (CH), 51.9 (CH_2), 63.9 (CH_2), 64.5 (CH_2, 68.7 (CH_2), 70.2 (CH_2), 81.3 (C_q), 117.9 (C_q), 128.1 (CH), 131.6 (CH) and 169.1 (C_q); *m/z* 354.2397 (M^+, 38%. $C_{20}H_{34}O_5$ requires 354.2406), 297 (42%), 239 (34), 195 (40), 153 (40), 99 (80) and 57 (100).

***tert*-Butyl [2-(3-oxo-2-pent-2-enylcyclopentyl)ethoxy]acetate 16**

Following the general procedure described above for compound **9**, the acetal **15** (340 mg, 0.96 mmol) was cleaved in the presence of THF and aq. HCl. The reaction mixture when flash chromatographed (MTBE–PE, 1:2) afforded **16** (232 mg, 0.75 mmol, 78%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1736 (ester and ketone); δ_H(400 MHz, $CDCl_3$) 0.92 (3H, t, *J* 7), 1.46 (9H, s), 1.5–1.58 (1H, m), 1.76–1.84 (1H, m), 1.97–2.1 (5H, m), 2.16–2.25 (1H, m), 2.27–2.35 (4H, m), 3.54–3.62 (2H, m), 3.92 (2H, s), 5.24 (1H, dtt, *J* 11, 7 and 1) and 5.40 (1H, dtt, *J* 11, 7 and 1); δ_C(100 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 25.3 (CH_2), 27.1 (CH_2), 28.0 (CH_3), 34.5 (CH_2), 37.9 (CH_2), 38.4 (CH), 54.9 (CH), 68.7 (CH_2), 69.5 (CH_2), 81.4 (C_q), 125.2 (CH), 133.5 (CH), 169.6 (C_q) and 220.1 (C_q); *m/z* 254 ($M^+ - Bu^t$, 24%), 236 (4), 209 (4), 186 (8), 151 (42), 148 (40), 106 (40) and 57 (100).

[2-(3-Oxo-2-pent-2-enylcyclopentyl)ethoxy]acetic acid 17: general procedure for the cleavage of *tert*-butyl esters

A solution of **16** (115 mg, 0.37 mmol) and trifluoroacetic acid (1.5 cm^3) in CH_2Cl_2 (3 cm^3) was stirred for 1 h at room temperature after which it was evaporated under reduced pressure. The residue was dissolved in aq. HCl and extracted with ethyl acetate. The extract was washed with water and brine, dried ($MgSO_4$) and concentrated by distillation under reduced pressure to give **17** (93 mg, 0.37 mmol, 100%) as a yellow oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3670 (OH) and 1733 (carboxy acid and ketone); δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, *J* 7), 1.38–1.50 (1H, m), 1.52–1.62 (1H, m), 1.81–1.88 (1H, m), 1.96–2.13 (5H, m), 2.16–2.24 (1H, m), 2.3–2.4 (3H, m), 3.65 (2H, t, *J* 7), 4.12 (2H, s), 5.24 (1H, dtt, *J* 11, 7 and 1) and 5.43 (1H, dtt, *J* 11, 7 and 1); δ_C(100 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 25.4 (CH_2), 27.1 (CH_2), 34.2 (CH_2), 38.0 (CH_2), 38.2 (CH), 54.9 (CH), 67.7 (CH_2), 69.9 (CH_2), 124.9 (CH), 133.9 (CH), 175.1 (C_q) and 221.8 (C_q); *m/z* 254 (M^+, 28%), 186 (20), 151 (52), 124 (8), 109 (16), 95 (25), 83 (52) and 69 (100).

Methyl [2-(3-oxo-2-pent-2-enylcyclopentyl)ethoxy]acetate 18: general procedure for the esterification with diazomethane

To **17** (72 mg, 0.28 mmol) was added dropwise a solution of diazomethane in Et_2O until the resulting solution remained yellow. After addition of aq. HCl, the mixture was extracted with MTBE. The extract was dried ($MgSO_4$) and evaporated under reduced pressure and the crude residue was chromatographed on silica gel (MTBE–PE, 1:2) to give **18** (64 mg, 0.24 mmol, 85%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1736 (ester and ketone); δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, *J* 8), 1.37–1.49 (1H, m), 152–1.60 (1H, m), 1.80–1.87 (1H, m), 1.97–2.12 (5H, m), 2.18–2.27 (1H, m), 2.3–2.39 (3H, m), 3.58–3.66 (2H, m), 3.76 (3H, m), 4.09 (2H, s), 5.26 (1H, dtt, *J* 11, 7 and 1) and 5.42 (1H, dtt, *J* 11, 7 and 1); δ_C(100 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 25.4 (CH_2), 27.1 (CH_2), 34.3 (CH_2), 37.9 (CH_2), 38.3 (CH), 51.7 (CH_3), 54.8 (CH), 68.1 (CH_2), 69.7 (CH_2), 125.2 (CH), 133.6 (CH), 170.8 (C_q) and 220.0 (C_q); *m/z* 268 (M^+, 22%), 200 (10), 151 (100), 131 (18), 110 (73), 95 (46), 83 (72) and 55 (66).

7-(2-Bromoethyl)-6-pent-2-enyl-1,4-dioxaspiro[4.4]nonane 19

To a solution of the alcohol **13** (1 g, 4.16 mmol), triphenylphosphine (1.37 g, 5 mmol) and pyridine (0.33 g) in CH_2Cl_2 (15 cm^3) was added a solution of CBr_4 (1.66 g, 5 mmol) over a period of 1 h. After being stirred for 2 h, the mixture was diluted with water (30 cm^3) and extracted with MTBE. The extract was dried ($MgSO_4$) and evaporated under reduced pressure. The crude residue was chromatographed on silica gel (MTBE–PE, 1:20) to give **19** (1.034 g, 3.4 mmol, 82%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3026 and 1205; δ_H(400 MHz, $CDCl_3$) 0.97 (3H, t, *J* 7.5), 1.23 (1H, dd, *J* 8 and 4.5), 1.6–1.9 (6H, m), 2.0–2.3 (5H, m), 3.35 (1H, dt, *J* 10 and 8), 3.43 (1H, ddd, *J* 5, 8 and 9), 3.82–3.90 (4H, m) and 5.31–5.43 (2H, m); *m/z* 302.0881 (M^+, 7%. $C_{14}H_{23}O_2Br$ requires 302.0875), 195 (25%), 153 (20), 114 (40), 99 (100) and 86 (38).

5-(6-Pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)pentan-1-ol 21

To a solution of the Grignard reagent **20**[11] (1 mmol) in dry THF (20 cm^3) at 0 °C under an atmosphere of Ar was added a 0.1 M solution of dilithium tetrachlorocuprate in dry THF (2 cm^3, 0.2 mmol) and a solution of **19** (200 mg, 0.66 mmol) in dry THF (2 cm^3). After 2 h, aq. NH_4Cl was added to the mixture which was then extracted with MTBE. The extract was dried ($MgSO_4$) and evaporated *in vacuo* and the residue was chromatographed on silica gel (MTBE–PE 1:1) to give **21** (141 mg, 0.05 mmol, 76%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3618 and 3464 (OH); δ_H(400 MHz, $CDCl_3$) 0.88 (3H, t, *J* 7), 1.05–1.32 (6H, m), 1.52–1.63 (4H, m), 1.66–1.87 (4H, m), 2.0–2.1 (3H, m), 2.17–2.25 (1H, m), 3.64 (2H, t, *J* 7), 3.82–3.93 (4H, m) and 5.31–5.43 (2H, m); *m/z* 282 (M^+, 10%), 253 (4), 220 (6), 195 (12), 153 (14), 114 (22), 99 (100), 86 (12), 67 (8) and 55 (8).

***tert*-Butyl [5-(6-pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)-pentyloxy]acetate 22**

Following the general procedure described above for compound **15**, the alcohol **21** (130 mg, 0.46 mmol) and *tert*-butyl bromoacetate **14** (400 mg, 2.1 mmol) were coupled. The reaction mixture when flash chromatographed (MTBE–PE, 1:1) afforded **22** (180 mg, 0.45 mmol, 98%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1742 (ester); δ_H(400 MHz, $CDCl_3$) 0.92 (3H, t, *J* 7), 1.0–1.18 (6H, m), 1.31 (9H, s), 1.42–1.53 (4H, m), 1.58–1.76 (4H, m), 1.9–2.0 (3H, m), 2.08–2.14 (1H, m), 3.48 (2H, t, *J* 6), 3.7–3.8 (4H, m), 3.86 (2H, s) and 5.28–5.37 (2H, m); *m/z* 396 (M^+, 20%), 339 (28), 295 (20), 271 (16), 195 (30), 183 (28), 153 (24), 40 (30), 99 (100) and 57 (56).

***tert*-Butyl [5-(3-oxo-2-pent-2-enylcyclopentyl)pentyloxy]acetate 23**

Following the general procedure described above for compound **9**, the acetal **22** (180 mg, 0.45 mmol) was cleaved in the presence of THF and aq. HCl. The reaction mixture when flash chromatographed (MTBE–PE, 1:2) afforded **23** (127 mg, 0.36 mmol, 80%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1741 (ester and ketone); δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, *J* 7), 1.19–1.41 (6H, m), 1.46 (9H, s), 1.54–1.83 (5H, m), 1.94–2.12 (4H, m), 2.24–2.4 (3H, m), 3.5 (2H, t, *J* 6), 3.95 (2H, s), 5.23 (1H, dtt, *J* 11, 7 and 1) and 5.41 (1H, dtt, *J* 11, 7 and 1); δ_C(100 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 25.3 (CH_2), 26.2 (CH_2), 26.9 (CH_2), 27.0 (CH_2), 28.0 (CH_3), 29.5 (CH_2), 34.6 (CH_2), 37.9 (CH_2), 41.0 (CH), 54.9 (CH), 68.7 (CH_2), 71.6 (CH_2), 81.4 (C_q), 125.4 (CH), 133.4 (CH), 169.7 (C_q) and 220.6 (C_q); *m/z* 296 ($M^+ - Bu^t$, 12%), 278 (22), 228 (30), 151 (60), 124 (32), 83 (62) and 57 (100).

[5-(3-Oxo-2-pent-2-enylcyclopentyl)pentyloxy]acetic acid 24
Following the general procedure described above for compound **17**, the ester **23** (144 mg, 0.41 mmol) was cleaved in the presence of trifluoroacetic acid. The crude product **24** (106 mg, 0.36 mmol, 88% yield) was used without further purification; $\nu_{max}(CHCl_3)/cm^{-1}$ 3164 (OH), 1735 (carboxy acid and ketone); δ_H(400 MHz, $CDCl_3$) 0.92 (3H, t, *J* 8), 1.20–1.50 (8H, m), 1.58–1.85 (6H, m), 1.96–2.16 (2H, m), 2.21–2.4 (2H, m), 3.57 (2H, t, *J* 7), 4.08 (2H, s), 5.23 (1H, dtt, *J* 11, 7 and 1), 5.41 (1H, dtt, *J* 11, 7 and 1); δ_C(100 MHz, $CDCl_3$) 14.1 (CH_3), 20.4 (CH_2), 25.3 (CH_2), 26.0 (CH_2), 26.7 (CH_2), 26.9 (CH_2), 29.3 (CH_2), 34.5 (CH_2), 37.9 (CH_2), 40.9 (CH), 54.9 (CH), 67.3 (CH_2), 71.8 (CH_2), 125.2 (CH), 133.4 (CH), 174.8 (C_q) and 221.4 (C_q); *m*/*z* 296 (M^+, 2%), 278 (8), 228 (12), 225 (20), 151 (32), 124 (28), 83 (86) and 66 (100).

Methyl [5-(3-oxo-2-pent-2-enylcyclopentyl)pentyloxy]acetate 25
Following the general procedure described above for compound **18**, the carboxy acid **24** (60 mg, 0.2 mmol) was esterified using diazomethane. The reaction mixture when flash chromatographed (MTBE–PE, 1:2) afforded **25** (50 mg, 0.16 mmol, 80%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1757 (ketone) and 1739 (ester); δ_H(400 MHz, $CDCl_3$) 0.94 (3H, t, *J* 8), 1.20–1.46 (8H, m), 1.52–1.85 (6H, m), 1.95–2.13 (2H, m), 2.25–2.41 (2H, m), 3.52 (2H, t, *J* 7), 3.75 (3H, s), 4.08 (2H, s), 5.23 (1H, dtt, *J* 11, 7 and 1) and 5.41 (1H, dtt, *J* 11, 7 and 1); δ_C(100 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 25.4 (CH_2), 26.2 (CH_2), 26.8 (CH_2), 27.0 (CH_2), 29.5 (CH_2), 34.6 (CH_2), 38.0 (CH_2), 41.0 (CH), 51.7 (CH_3), 55.0 (CH), 68.2 (CH_2), 71.8 (CH_2), 125.4 (CH), 133.4 (CH), 170.9 (C_q) and 220.6 (C_q); *m*/*z* 310 (M^+, 12%), 292 (32), 242 (36), 151 (96), 124 (80), 95 (80) and 83 (100).

2-{2-(2-Pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)ethoxy}-ethanol 26
A solution of **15** (1 g, 2.6 mmol) in THF (20 cm^3) was added to a suspension of $LiAlH_4$ (0.15 g, 4 mmol) in dry THF (30 cm^3) and the mixture was stirred for 2 h at room temperature. After that, it was cooled to 0 °C and treated with ice–water until hydrogen evolution stopped. The precipitate was filtered off and the filtrate was extracted with MTBE. The combined extracts were dried ($MgSO_4$) and evaporated *in vacuo*. Flash chromatography of the residue on silica gel (MTBE–PE, 1:1) afforded **26** (665 mg, 2.3 mmol, 89%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3625 (OH), 3474 (OH), 3030 and 1039; δ_C(400 MHz, $CDCl_3$) 0.96 (3H, t, *J* 7.5), 1.47 (1H, dddd, *J* 5.5, 7, 9 and 13), 1.6–1.91 (7H, m), 2.05 (2H, m), 2.07 (1H, dd, *J* 5.5 and 7.7), 2.24 (1H, m), 3.58–3.72 (2H, m), 3.82–3.93 (4H, m) and 5.38 (2H, m); *m*/*z* 284.1988 (M^+, 38%. $C_{16}H_{28}O_4$ requires 284.1980), 195 (34%), 153 (60), 114 (24) and 99 (100).

***tert*-Butyl {2-[2-(6-pent-2-enyl-1,4-dioxaspiro[4.4]nonan-7-yl)-ethoxy]ethoxy}acetate 27**
Following the general procedure described for compound **15**, the alcohol **26** (350 mg, 1.2 mmol) and *tert*-butyl bromoacetate **14** (730 mg, 3.7 mmol) were coupled. The reaction mixture when flash chromatographed (MTBE–PE, 1:4) afforded **27** (378 mg, 0.9 mmol, 79%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 3014, 2966, 1743, 1230 and 1042; δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, *J* 8), 1.26 (2H, m), 1.47 (9H, s), 1.75 (5H, m), 1.92 (1H, m), 2.05 (3H, m), 2.22 (1H, ddd, *J* 5.5, 6.5 and 14), 3.41 (2H, m), 3.6 (2H, m), 3.7 (2H, m), 3.85 (4H, m), 4.02 (2H, s) and 5.36 (2H, m); *m*/*z* 398 (M^+, 20%), 342 (56), 280 (16), 239 (16), 195 (44), 153 (40), 99 (100) and 57 (54).

***tert*-Butyl {2-[2-(3-oxo-2-pent-2-enylcyclopentyl)ethoxy]-ethoxy}acetate 28**
Following the general procedure described above for compound **9**, the acetal **27** (320 mg, 0.8 mmol) was cleaved in the presence of THF and aq. HCl. The reaction mixture when flash chromatographed (MTBE–PE, 1:2) afforded **28** (273 mg, 0.77 mmol, 96%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 2965 and 1735; δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, *J* 8), 1.42 (9H, s), 1.57 (4H, m), 1.72 (1H, m), 1.85 (1H, m), 2.0–2.2 (6H, m), 2.35 (2H, m), 3.57 (2H, m), 3.62 (2H, m), 4.02 (2H, s), 5.25 (1H, dtt, *J* 10.5, 7.5 and 1.5) and 5.42 (1H, dtt, *J* 10.5, 7.5 and 1.5); *m*/*z* 298.178 ($M^+ - C_4H_8$, 16%. $C_{16}H_{26}O_5$ requires 298. 1773), 280 (8%), 253 (12), 205 (6), 151 (26), 81 (54) and 73 (100).

{2-[2-(3-Oxo-2-pent-2-enylcyclopentyl)ethoxy]ethoxy}acetic acid 29
Following the general procedure described above for compound **17**, the ester **28** (200 mg, 0.67 mmol) was cleaved in the presence of trifluoroacetic acid. The crude product **29** (145 mg, 0.46 mmol, 69%) was used without further purification; $\nu_{max}(CHCl_3)/cm^{-1}$ 1736 (ketone), 1706 (carboxy acid) and 1228; δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, *J* 7.5), 1.45 (1H, ddt, *J* 7.5, 13.5 and 10.5), 1.62 (1H, ddt, *J* 9, 13.5 and 6), 1.85 (1H, m), 1.95–2.25 (6H, m), 2.36 (3H, m), 3.63 (4H, m), 3.77 (2H, dd, *J* 5 and 3), 4.17 (2H, s), 5.25 (1H, dtt, *J* 10, 7 and 1.5) and 5.43 (1H, dtt, *J* 10, 7 and 1.5); δ_C(67.5 MHz, $CDCl_3$) 14.1 (CH_3), 20.5 (CH_2), 25.3 (CH_2), 27.1 (CH_2), 34.2 (CH_2), 37.9 (CH_2), 38.2 (CH), 35.4 (CH_2), 68.4 (CH_2), 69.5 (CH_2), 69.8 (CH_2), 71.1 (CH_2), 125.1 (CH), 133.6 (CH), 173.7 (C_q) and 220.5 (C_q); *m*/*z* 298 (M^+, 7%), 284 (6), 205 (12), 149 (40), 110 (34), 99 (80) and 73 (100).

Methyl {2-[2-(3-oxo-2-pent-2-enylcyclopentyl)ethoxy]ethoxy}-acetate 30
Following the general procedure described above for compound **18**, the carboxy acid **29** (70 mg, 0.23 mmol) was esterified using diazomethane. The reaction mixture when flash chromatographed (MTBE–PE, 1:1) afforded **30** (66 mg, 0.21 mmol, 83%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1735 (ester and ketone), 1232 and 1149; δ_H(400 MHz, $CDCl_3$) 0.95 (3H, t, *J* 7.5), 1.42 (2H, m), 1.55 (1H, ddd, *J* 13, 7 and 2), 1.82 (1H, m), 1.9–2.2 (5H, m), 2.35 (3H, m), 3.55 (2H, m), 3.65 (2H, m), 3.7 (4H, m), 4.17 (3H, s), 5.25 (1H, dtt, *J* 11, 7 and 1.5) and 5.42 (1H, dtt, *J* 11, 7 and 1.5); δ_C(67.5 MHz, $CDCl_3$) 14.0 (CH_3), 20.4 (CH_2), 25.2 (CH_2), 27.0 (CH_2), 34.2 (CH_2), 35.4 (CH_2), 37.8 (CH_2), 38.2 (CH), 51.6 (CH_3), 68.4 (CH_2), 69.2 (CH_2), 70.1 (CH_2), 70.7 (CH_2), 125.1 (CH), 133.4 (CH), 170.7 (C_q) and 220.1 (C_q); *m*/*z* 312.1937 (M^+, 7%. $C_{17}H_{28}O_5$ requires 312.1929), 284 (2%), 247 (3), 178 (24), 149 (50), 110 (62) and 55 (100).

Dimethyl 2-(3-oxo-2-pent-2-ynylcyclopentyl)malonate 32
To a solution of Na (7 mg, 0.3 mmol) in dry methanol (10 cm^3) was added dimethyl malonate (574 mg, 4.35 mmol) at room temperature. After 10 min the solution was cooled to 0 °C and **31** (425 mg, 2.9 mmol) was added to it; stirring was continued at the same temperature for 2 h. The mixture was then quenched with 1 M aq. HCl (20 cm^3) and extracted with MTBE. The extent was washed with brine, dried ($MgSO_4$) and concentrated by distillation under reduced pressure to give the crude product **32** (450 mg, 1.6 mmol, 55%). The crude product was used in the next reaction without purification; $\nu_{max}(CHCl_3)/cm^{-1}$ 1738 (ketone and ester), 1160 and 1077 (s); δ_H(400 MHz, $CDCl_3$) 1.09 (3H, t, *J* 7), 1.73–1.85 (1H, m), 2.01–2.3 (4H, m), 2.15 (2H, qt, *J* 7 and 2.5), 2.36–2.59 (3H, m), 2.81–2.91 (1H, m), 3.76 (3H, s) and 3.78 (3H, s); *m*/*z* 251 ($M^+ - C_2H_5$, 26%), 189 (7), 161 (6), 148 (90), 133 (30), 122 (23), 107 (16), 85 (30), 71 (54) and 57 (100).

Methyl (3-oxo-2-pent-2-ynylcyclopentyl)acetate 33
A mixture of the crude diester **32** (335 mg, 1.2 mmol) and adipic acid (140 mg, 1.0 mmol) was heated at 190 °C for 6 h after which it was dissolved in MTBE (50 cm^3) and the solution washed with aq. $NaHCO_3$. The organic layer was dried ($MgSO_4$) and evaporated *in vacuo* and the residue was chromatographed on silica gel (MTBE–PE, 1:4) to give **33** (55 mg, 0.25 mmol, 21%) as a colourless oil; $\nu_{max}(CHCl_3)/cm^{-1}$ 1736 (ketone and ester); δ_H(400 MHz, $CDCl_3$) 1.08 (3H, t, *J* 8), 1.45–

1.55 (1H, m), 1.9–1.96 (1H, m), 2.11 (2H, qt, *J* 8 and 3), 2.23–2.34 (2H, m), 2.34–2.43 (2H, m), 2.46–2.5 (2H, m), 2.5–2.61 (1H, m), 2.85 (1H, dd, *J* 15 and 4.5) and 3.71 (3H, s); δ_C(100 MHz, $CDCl_3$) 12.2 (CH_2), 14.0 (CH_3), 17.3 (CH_2), 27.0 (CH_2), 37.5 (CH_2), 37.8 (CH), 38.4 (CH_2), 51.5 (CH_3), 52.7 (CH), 75.6 (CH), 83.6 (CH), 172.4 (C_q) and 217.3 (C_q); *m/z* 222 (M^+, 0.7%), 207 (2), 193 (42), 149 (12), 133 (14), 122 (100), 107 (50), 91 (27), 79 (25), 67 (10) and 55 (14).

Methyl [6-(2-oxoethyl)-1,4-dioxaspiro[4.4]nonan-7-yl]acetate 34
A solution of **7** (100 mg, 0,37 mmol) in CH_2Cl_2 (100 mg, 0.37 mmol) was ozonized at −78 °C, the reaction being stopped when the mixture became blue. The excess of ozone was flushed from the mixture with N_2 after which it was slowly added to a stirred suspension of Zn powder (300 mg) in acetic acid (3 cm^3). The mixture was then washed with aq. $NaHCO_3$, water and brine, and dried ($MgSO_4$) to give aldehyde **34** (73 mg, 0.3 mmol, 81%) as a colourless oil; ν_{max}($CHCl_3$)/cm^{-1} 1730 (ester and aldehyde); δ_H(400 MHz, $CDCl_3$) 1.38 (1H, ddt, *J* 13, 9 and 9), 1.7–1.86 (2H, m), 1.9–2.01 (1H, m), 2.09–2.2 (1H, m), 2.23–2.4 (3H, m), 2.49–2.62 (2H, m), 3.65 (3H, s), 3.78–3.95 (4H, m) and 9.73 (1H, m); δ_C(50 MHz, $CDCl_3$) 28.0 (CH_2), 34.7 (CH_2), 38.9 (CH), 39.2 (CH_2), 43.0 (CH_2), 46.7 (CH), 51.6 (CH_3), 64.3 (CH_2), 64.7 (CH_2), 117.0 (C_q), 173.0 (C_q) and 202.0 (C_q); *m/z* 242 (M^+, 3%), 213 (30), 185 (18), 169 (16), 99 (100) and 55 (24).

Methyl [6-(4-hydroxybut-2-enyl)-1,4-dioxaspiro[4.4]nonan-7-yl]acetate 36
To a stirred suspension of the phosphonium salt **35** (2.5 g, 6.5 mmol) in dry THF (30 cm^3) was added a solution of butyllithium in hexane (1.6 M; 8.2 cm^3, 13.2 mmol) at 0 °C. The resulting red solution was cooled to −30 °C and treated with a solution of the aldehyde **34** (1.045 g, 4.35 mmol) in dry THF (10 cm^3). After decolorization of the solution stirring was continued at −30 °C for a further 2 h. After addition of aq. NH_4Cl, the mixture was extracted with MTBE. The extract was dried ($MgSO_4$) and the evaporated *in vacuo* and the residue was chromatographed on silica gel (MTBE–PE, 1 : 2) to give **36** (200 mg, 0.74 mmol, 17%) as a 2 : 1 mixture of *E*/*Z*-isomers (colourless oil); ν_{max}($CHCl_3$)/cm^{-1} 3500 (OH) and 1733 (ester); δ_H(400 MHz, $CDCl_3$) 1.24–1.37 (1H, m), 1.66–2.34 (9H, m), 2.49–2.58 (1H, m), 3.66 (3H, s), 3.82–3.94 (4H, m), 4.05–4.10 (1.33H, m, *E*), 4.16–4.21 (0.66H, m, *Z*) and 5.55–5.70 (2H, m); *m/z* 270.1467 (M^+, 1%. $C_{14}H_{22}O_5$ requires 270.1467), 252 (20%), 197 (10), 155 (12), 116 (10), 99 (100), 86 (16) and 55 (10).

Methyl [2-(4-hydroxybut-2-enyl)-3-oxocyclopentyl]acetate 37a/b
Following the general procedure described above for compound **9**, the acetal **36**, (150 mg, 0.56 mmol) was cleaved in the presence of THF and aq. HCl. After work-up, the crude residue was chromatographed on Sephadex (CH_2Cl_2) to give the *E*-isomer of **37b** (82 mg, 0.36 mmol, 64%) and the *Z*-isomer of **37a** (42 mg, 0.19 mmol, 34%) as colourless oils; ν_{max}($CHCl_3$)/cm^{-1} 3520 (OH) and 1735 (ester and ketone); *m/z* 208.1099 ($M^+ - H_2O$, 56%. $C_{12}H_{16}O_3$ requires 208.1099), 193 (42%), 167 (22), 135 (92), 117 (30), 93 (44), 83 (100) and 79 (55); *Z*-isomer: δ_H(400 MHz, $CDCl_3$) 1.51 (1H, m), 1.94 (1H, m), 2.05–2.48 (7H, m), 2.45–2.53 (1H, m), 2.66 (1H, dd, *J* 11 and 5), 3.71 (3H, s), 4.17 (2H, dd, *J* 7 and 5), 5.45 (1H, dtt, *J* 11, 7 and 1.5) and 5.74 (1H, dtt, *J* 11, 7 and 1.5); δ_C(50 MHz, $CDCl_3$) 25.0 (CH_2), 27.4 (CH_2), 37.6 (CH_2), 37.8 (CH), 38.8 (CH_2), 51.8 (CH_3), 54.1 (CH), 58.1 (CH_2),128.4 (CH), 131.0 (CH), 172.8 (C_q) and 219.0 (C_q); *E*-isomer: δ_H(400 MHz, $CDCl_3$) 1.57 (1H, m), 1.97 (1H, m), 2.11 (1H, m), 2.20–2.27 (1H, m), 2.27–2.4 (6H, m), 2.65 (1H, m), 3.7 (3H, s), 4.08 (2H, t, *J* 5), 5.6 (1H, dtt, *J* 15, 7 and 1) and 5.71 (1H, dtt, *J* 15, 5 and 1); δ_C(50 MHz, $CDCl_3$) 27.1 (CH_2), 30.5 (CH_2), 37.7 (CH_2), 37.6 (CH), 38.6 (CH_2), 51.7 (CH_3), 53.8 (CH), 63.2 (CH_2), 128.6 (CH), 132.1 (CH), 172.5 (C_q) and 218.6 (C_q).

Methyl {2-[10-(2-methoxycarbonylmethyl-5-oxocyclopentyl)-deca-2,8-dienyl]-3-oxocyclopentyl}acetate 40
To a stirred suspension of the phosphonium salt **39** (760 mg, 1.0 mmol) in dry THF (15 cm^3) was added a solution of butyllithium in hexane (1.6 M; 1.2 cm^3, 2 mmol) at 0 °C. The resulting red solution was cooled to −30 °C and a solution of the aldehyde **38** (352 mg, 1.8 mmol) in dry THF (10 cm^3) was added to it. After decolorization of the solution stirring was continued at −30 °C for a further 2 h. The mixture was then diluted with aq. NH_4Cl and extracted with MTBE. The extract was dried ($MgSO_4$) and evaporated *in vacuo* and the residue was chromatographed on silica gel (MTBE–PE, 1 : 5) to give **40** (50 mg, 0.11 mmol, 12%) as a colourless oil; ν_{max}($CHCl_3$)/cm^{-1} 1743 (ester and ketone); δ_H(200 MHz, $CDCl_3$) 1.24–1.38 (4H, m), 1.42–1.59 (2H, m), 1.80–2.43 (20H, m), 2.62–2.78 (2H, m), 3.68 (6H, s), 5.28 (2H, dtt, *J* 11, 7 and 1.5) and 5.44 (2H, dtt, *J* 11, 7 and 1.5); δ_C(50 MHz, $CDCl_3$) 25.6 (CH_2), 27.2 (CH_2), 29.2 (CH_2), 37.7 (CH_2), 38.0 (CH), 38.8 (CH_2), 51.6 (CH_3), 54.0 (CH), 125.7 (CH), 132.2 (CH), 172.5 (C_q) and 218.9 (C_q); *m/z* 446.2668 (M^+, 10%. $C_{13}H_{38}O_6$ requires 446.2668), 373 (6%), 341 (6), 279 (32), 265 (20), 167 (52), 149 (100), 107 (20), 97 (42), 83 (40) and 57 (30).

Methyl {6-[4-(7-methoxycarbonylmethyl-1,4-dioxaspiro[4.4]-non-6-yl)but-2-enyl]-1,4-dioxaspiro[4.4]nonan-7-yl}acetate 42
A solution of **41** (64 mg, 0.27 mmol) and **[Mo]** (5 mol%; 10 mg, 0.013 mmol) in CH_2Cl_2 (3 cm^3) was refluxed for 4 h in a glove box under an atmosphere of argon. The reaction mixture when flash chromatographed (MTBE–PE, 1 : 1) afforded **42** (53 mg, 0.12 mmol, 89%) as a 2 : 1 mixture of *E*/*Z*-isomers (colourless oil). The configuration of the double bond produced was determined by NMR and GC methods; ν_{max}(ATR)/cm^{-1} 1734 (ester); δ_H(200 MHz, $CDCl_3$) 1.15–1.38 (2H, m), 1.55–1.79 (6H, m), 1.81–2.35 (12H, m), 2.49–2.64 (2H, m), 3.63 (6H, s), 3.80–3.92 (8H, m) and 5.37–5.47 (2H, m); δ_C(50 MHz, $CDCl_3$) 27.9 (CH_2), 32.1 (CH_2), 35.1 (CH_2), 33.9 (CH_2), 38.9 (0.67 CH, *E*), 39.3 (0.33 CH, *Z*), 40.0 (CH_2), 51.0 (CH), 51.3 (CH_3), 64.1 (CH_2), 64.6 (CH_2), 117.6 (C_q), 128.9 (0.67 CH, *E*), 130.0 (0.33 CH, *Z*) and 173.2 (C_q); *m/z* 452.241 (M^+, 4%. $C_{24}H_{36}O_8$ requires 452.2410), 421 (8%), 390 (4), 337 (4), 298 (4), 252 (66), 225 (8), 200 (40), 169 (10), 149 (10), 127 (24), 99 (100), 86 (12), 55 (10) and 43 (12).

Methyl {2-[4-(2-methoxycarbonylmethyl-5-oxocyclopentyl)but-2-enyl]-3-oxocyclopentyl}acetate 43
A solution of **42** (57 mg, 0.126 mmol), pyridinium tosylate (2 mg), water (1 cm^3) and acetone (2 cm^3) was stirred at 60 °C for 2 h. Excess of solvent was removed *in vacuo* from the mixture which was then diluted with MTBE (10 cm^3), washed with sat. aq. $NaHCO_3$, water and brine, dried ($MgSO_4$) and evaporated under reduced pressure to give pure **43** (36 mg, 0.1 mmol, 80%) as a colourless oil; ν_{max}(ATR)/cm^{-1} 1733 (ketone and ester); δ_H(200 MHz, $CDCl_3$) 1.43–1.55 (2H, m), 1.82–1.96 (2H, m), 2.04–2.16 (2H, m), 2.17–2.43 (12H, m), 2.57–2.74 (2H, m), 3.69 (6H, s) and 5.33–5.45 (2H, m); δ_C(50 MHz, $CDCl_3$) 27.1 (CH_2), 30.5 (CH_2), 37.3 (CH), 37.7 (0.33 CH_2, *Z*), 37.9 (0.67 CH_2, *E*), 38.5 (0.67 CH_2, *E*), 38.6 (0.33 CH_2, *Z*), 51.5 (CH_3), 53.9 (CH), 128.1 (0.33 CH, *Z*), 129.2 (0.67 CH, *E*), 172.4 (C_q) and 218.6 (C_q); *m/z* 364.1886 (M^+, 40%. $C_{20}H_{28}O_6$ requires 364.1886), 333 (20%), 291 (16), 259 (20), 208 (100), 193 (27), 177 (54), 156 (90), 135 (48), 117 (22), 91 (26), 83 (96), 67 (16) and 55 (12).

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