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Synthesis of volicitin: a novel three-component Wittig approach to chiral 17-hydroxylinolenic acid

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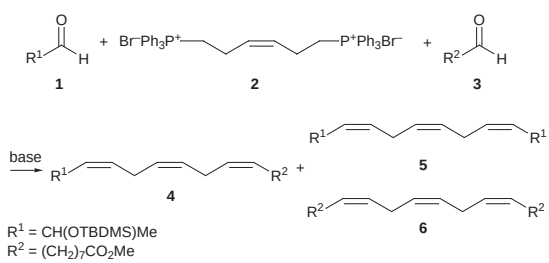
A short stereoselective synthesis of both enantiomers of *N*-(17-hydroxylinolenoyl)-L-glutamine (volicitin) 12, an elicitor of plant volatile biosynthesis from beet armyworm salivary secretion, is described; the protected 17-hydroxylinolenic acid 4 moiety of volicitin 12 was prepared in a one pot bis-Wittig reaction.

Plants are able to respond to herbivore damage by *de novo* biosynthesis of a characteristic blend of volatiles that attracts the natural enemies of the attacking insect.[1] Feeding on the leaf is accompanied by introduction of salivary secretions that may contain high and/or low molecular weight compounds activating the plant's defense. Established elicitors are a β-glucosidase from the regurgitant of *Pieris brassicae* caterpillars[2] and the recently identified *N*-(17-hydroxylinolenoyl)-L-glutamine (volicitin) **12** from beet armyworm oral secretion.[3] Like the microbial metabolite coronatine, which strongly induces volatile biosynthesis in plants,[4] volicitin **12** is also an amphiphilic compound that consists of a nonpolar lipid moiety joined to an amino acid by an amide linkage. First experiments with synthetic volicitin revealed that biological activity is highly dependent on the stereochemistry of the amino acid.[3] A conjugate of (±)-17-hydroxylinolenic acid and L-glutamine proved to be highly active, while the corresponding conjugate with D-glutamine was virtually inactive. The configuration of the 17-hydroxy group of the fatty acid has not been addressed to date.

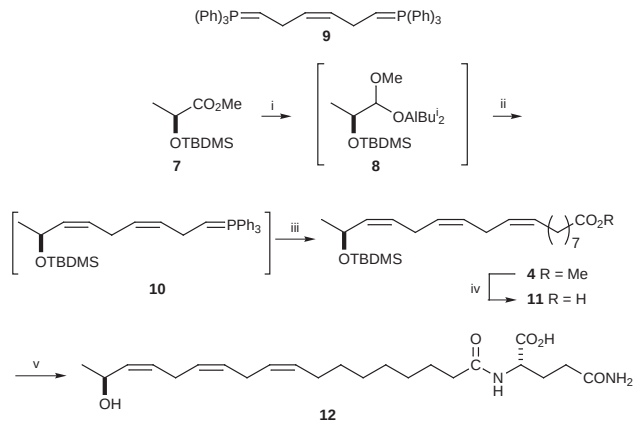
Here we report an efficient synthesis of both enantiomers of TBDMS protected 17-hydroxylinolenic acid methyl ester **4** based on a novel three component bis-Wittig approach. Subsequent deprotection and coupling with L-glutamine yielded both volicitin diastereomers (17*R*)- and (17*S*)-**12**, respectively.

According to Scheme 1, the fatty acid skeleton is readily assembled in a single operation *via* a bis-Wittig approach, based on the *Z*-hex-3-ene-1,6-diol-derived symmetric bifunctional Wittig salt **2**.[5] In order to suppress the formation of symmetric by-products **5** and **6**, resulting from statistical coupling of the three components **1**,[6] **2** and **3**, the reaction conditions were carefully optimised. While addition of a 1 : 1 mixture of the aldehydes **1** and **3** to the ylide **9** resulted in the expected statistical product ratio of **4**, **5** and **6**, successive addition of the aldehydes to **9**, in conjunction with optimised temperatures during the stepwise reaction, afforded high yields of the protected 17-hydroxylinolenic acid **4**. This result suggested



Scheme 1

different reactivity of the bis-functional ylide **9** and the monofunctional intermediates like **10**. Thus, if only 1 equiv. of aldehyde **1** was added to a cold solution (−100 °C) of the bis-ylide **9**, generated from the bis-phosphonium salt with 2.1 equiv. of KHMDS,[7] a preferential formation of the mono-ylide **10** was observed after slow warming to −20 °C. Recooling to −78 °C and addition of 1 equiv. of aldehyde **3** afforded the cross-coupled product **4** in high yield. According to GLC-MS analysis, the amount of the two symmetric products **5** and **6** was less than 20% of the product mixture. Furthermore, the amount of symmetric coupling products could be suppressed even more efficiently by using the aluminium alkoxide **8**, generated by reduction of TBDMS-protected D- or L-lactic acid methyl ester **7** with DIBAL-H at low temperature, as the first carbonyl equivalent (Scheme 2).[8]



Scheme 2 *Reagents and conditions*: i, DIBAL-H, Et_2O, −78 °C; ii, **9** [prepared from **2** in THF using 2 equiv. $KN(SiMe_3)_2$, −78 to 0 °C], −78 °C, add **8**, warm to room temp.; iii, −78 °C, **3**, warm to room temp.; iv, LiOH, THF–H_2O; v, NEt_3, THF, $ClCO_2Et$, −10 °C, then Gln, NaOH, room temp.

During warming to room temperature, the silylated aldehyde **1** is presumably slowly released from the intermediate aluminium complex **8** and reacts instantaneously with the bis-ylide **9** to give the ylide **10**. After 30 min at room temperature, the solution was recooled (−78 °C), before 1.2 equiv. of the 9-oxo ester **3** were added. Following work-up, the silylated (17*R*)- or (17*S*)-hydroxylinolenic acid **4** could be isolated in 42% overall yield.† Less than 10% of the symmetrical coupling products **5** and **6** could be detected. Use of the configurationally stable aluminium complex **8** had the additional advantage that the sensitive chiral C_3 synthon **1** reacted without racemisation under the basic Wittig conditions. Both products, (17*S*)- and (17*R*)-**4**, proved to be of >97% ee by Mosher ester analysis of the end products (17*S*)- and (17*R*)-**12** (volicitin).‡ ^{13}C NMR analysis revealed the configuration of the 9- and 15-double bonds to be of >95% *Z*.

Saponification of **4** with aqueous LiOH in THF[7] afforded TBDMS protected 17-hydroxylinolenic acid **11** in 86% yield. Coupling of **11** with glutamine was achieved using the ethyl carbonate mixed anhydride method. Acidic workup of the crude

product resulted in simultaneous deprotection, and pure volicitin **4** could be separated from excess glutamine by RP-MPLC-chromatography.§ The significance of the chiral centre for the induction of plant volatile biosynthesis is currently being evaluated. Detailed results together with information on the mode of signaling of the compound will be presented elsewhere.

The three-component Wittig reaction described here opens a new and highly versatile route to the skipped triene substructures occurring in a large variety of natural products. The central building block **2** is readily available on a multi-gram scale,[5] and one-pot product formation with good stereoselectivities allows its universal use as a building block in the synthesis of highly unsaturated fatty acids and related compounds. Like the previously introduced di-n-butyl-1-stannacyclohexa-2,5-diene[9] as a bifunctional building block for the formation of skipped dienes, the bis-Wittig approach also has universal potential, allowing the direct assembly of at least three homoconjugated double bonds from simple precursors in a single operation.

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Notes and references

† Experimental procedure: A cold (−78 °C) suspension of the Wittig salt **2** (700 mg, 0.91 mmol) in dry THF (15 ml) was treated with $KN(SiMe_3)_2$ in hexane (3.8 ml of a 0.5 M solution). The solution was allowed to warm to 0 °C, stirred for 10 min and recooled (−78 °C). An ethereal solution of the aluminate **8** (2.3 ml, 0.4 M, pre-cooled to −78 °C) was added (ref. 6,8). The mixture was allowed to warm to room temperature and stirring was continued for 1 h before recooling to −78 °C. Then, a solution of **3** (203 mg in 1 ml THF) was added and the mixture was allowed again to reach room temperature. Hydrolysis with sat. NH_4Cl, extraction with Et_2O and column chromatography on SiO_2 yielded the protected ester **4** as a colourless, viscous liquid. Yield: 161 mg (42%).

‡ Both enantiomers of **12** were esterified with excess (*R*)-(−)-α-methoxy-α-(trifluoromethyl)phenylacetyl chloride in the presence of DMAP and Et_3N (ref. 10). After the reaction was complete (no starting material could be detected *via* NMR analysis) the ee of **12** was determined by integration of the isolated 17-hydroxylinolenic acid C(18)-methyl doublet [δ 1.35 for (17*S*)-**12** and δ 1.42 for (17*R*)-**12**].

§ *Selected data* for **12**: $[\alpha]_{589}^{22}$ +3 (*c* 0.83, CH_2Cl_2) [(17*S*)-**12**]; $[\alpha]_{589}^{22}$ −4.0 (*c* 0.82, CH_2Cl_2) [(17*R*)-**12**]; δ_H (CD_3OD, 500 MHz) 5.3–5.17 (m, 6H), 4.50 (dq, 1H, *J* 6.8, 6.36), 4.25 (dd, 1H, *J* 4.8, 3.9), 2.75 (m, 2H), 2.69 (t, 2H, *J* 5.8), 2.21–2.16 (m, 2H), 2.12 (t, 2H, *J* 7.3), 2.07–1.95 (m, 1H), 1.95 (q, 2H, *J* 6.5), 1.87–1.77 (m, 1H), 1.49 (t, 2H), 1.29–1.18 (m, 10H), 1.07 (d, 3H, *J* 6.36); δ_C (CD_3OD, 125 MHz), 176.761, 175.404, 174.169, 134.418, 130.237, 128.657, 128.190, 127.690, 127.680, 63.396, 52.376, 35.867, 31.799, 29.704, 29.339, 29.269, 29.227, 27.623, 27.177, 25.868, 25.787, 25.519, 22.970. See ref. 3 for mass spectroscopic data.

1 M. K. Stowe, T. C. J. Turlings, J. H. Loughrin, W. J. Lewis and J. H. Tumlinson, *Proc. Natl. Acad. Sci. U.S.A.*, 1995, **92**, 23 and references cited therein.
2 M. L. Mattiacci, M. Dicke and M. A. Posthumus, *Proc. Natl. Acad. Sci. U.S.A.*, 1995, **92**, 2036.
3 H. T. Alborn, T. C. J. Turlings, T. H. Jones, G. Stenhagen, J. H. Loughrin and J. H. Tumlinson, *Science*, 1997, **276**, 945.
4 W. Boland, J. Hopke, J. Donath, J. Nüske and F. Bublitz, *Angew. Chem., Int. Ed. Engl.*, 1995, **34**, 1600.
5 P. Fiedler, Dissertation, Universität Karlsruhe (TH), 1980. Experimental details for the preparation of **2** will be reported elsewhere.
6 S. K. Massad, L. D. Hawkins and D. C. Baker, *J. Org. Chem.*, 1983, **48**, 5180.
7 K. C. Nicolaou, J. Y. Ramphal and Y. Abe, *Synthesis*, 1989, 898.
8 W. Boland, P. Ney and L. Jaenicke, *Synthesis*, 1980, 1015.
9 E. J. Corey, J. Kang, *Tetrahedron Lett.*, 1982, **23**, 1651.
10 J. A. Dale, D. L. Dull and H. S. Mosher, *J. Org. Chem.*, 1969, **34**, 2543.

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