Available online at www.sciencedirect.com
SCIENCE DIRECT®
Tetrahedron Letters 45 (2004) 7851–7853
Tetrahedron Letters

Synthesis and protein reactivity of 2*E*,4*E*,6*E*-dodecatrienal

Chongzhao Ran, Weihan Zhang and Lawrence M. Sayre*

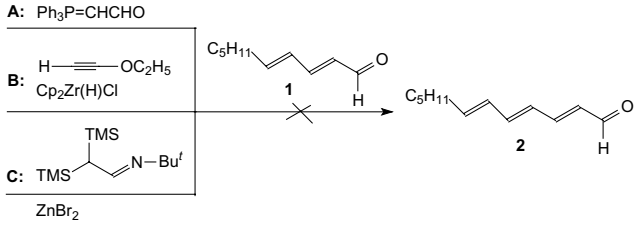
Department of Chemistry, Case Western Reserve University, Cleveland, OH 44106, USA

Received 12 August 2004; revised 26 August 2004; accepted 26 August 2004

Abstract—An efficient approach for synthesis of the food odorant 2*E*,4*E*,6*E*-dodecatrienal (DTE) by extension of 2*E*,4*E*-decadienal (DDE) is reported. DTE shows higher protein crosslinking ability than the lipid peroxidation products DDE and 4-hydroxy-2*E*-nonenal.

2*E*,4*E*,6*E*-Dodecatrienal (DTE) **2** is a component of the volatile fraction of soybean oil[1] and roasted beef fat[2] that contributes to their characteristic aroma, and is a major constituent of the volatile fraction of roasted sesame seeds.[3] Although the biological properties of DTE have not been investigated, the lower enals (2-enals, and 2,4-dienals) have been studied extensively as cytotoxic reactive electrophilic products of lipid peroxidation that are capable of modifying DNA and proteins. For example, 2*E*,4*E*-decadienal (DDE) strongly inhibits human erythroleukemia cell growth and is involved in the beginning of DNA fragmentation.[4] DDE modifies DNA bases[5] and was found to crosslink human erythrocyte ghost proteins.[6] We recently found that DDE, along with other common lipid oxidation products, 4-hydroxy-2-nonenal (HNE) and 4-oxo-2-nonenal (ONE), are capable of crosslinking protein[7] and crosslinking DNA and associated histone protein.[8] We wondered if the reactivity of such compounds would extend to the trienal series. For this reason, we synthesized DTE by an efficient approach that should be applicable to the preparation of other aliphatic polyene aldehydes. In addition, we determined the protein crosslinking ability of DTE relative to HNE, ONE, and DDE.

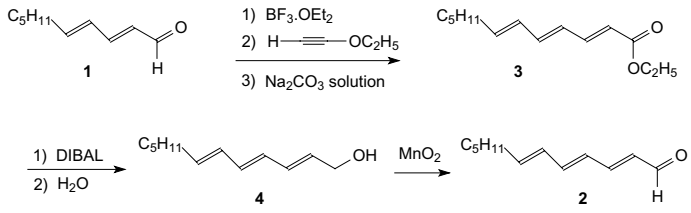
To synthesize DTE (**2**) from commercial available DDE (**1**), several reported approaches (Scheme 1) were first tried. The most obvious approach was a Wittig elongation (Scheme 1A), and the required Wittig reagent was commercial available. However, the reaction failed under a variety of reaction conditions, possibly because **1** is an *aliphatic* polyene aldehyde, and the Wittig elongation has been reported only for *aryl*polyene aldehydes.[9] It was also reported that aliphatic polyene aldehydes could be synthesized by two carbon homologation with ethyl ethynyl ether, mediated by Schwartz's reagent ($Cp_2Zr(H)Cl$) with a catalytic amount of $AgClO_4$ in dichloromethane (Scheme 1B).[10] When this reaction was carried out, the hydrozirconation appeared to proceed smoothly, but no product could be recovered from the subsequent step with aldehyde **1** in the presence of $AgClO_4$. A more delicate approach to prepare polyene aldehydes relies on the use of *N*-*tert*-butylacetaldimine to prepare α,α-bis(trimethylsilyl)-*tert*-butylacetaldimine as a precursor, which then reacts with polyene aldehydes using $ZnBr_2$ as catalyst (Scheme 1C).[11] In our hands we could not obtain **2** by this method. Lastly, in a report investigating whether **2** might be a product of autoxidation of arachidonic acid (it is not),[12] a reference amount of **2** was prepared by microsynthesis via aldol condensation of acetaldehyde and **1**, though the yield was not mentioned. We failed to obtain **2** from **1** on a preparative scale using this method under a variety of reaction conditions.



Scheme 1.

Keywords: Dodecatrienal; Decadienal; Bifunctional aldehyde; Protein crosslinking.
* Corresponding author. Tel.: +1 2163683704; fax: +1 2163683006; e-mail: lms3@case.edu

0040-4039/$ - see front matter © 2004 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tetlet.2004.08.158



Scheme 2.

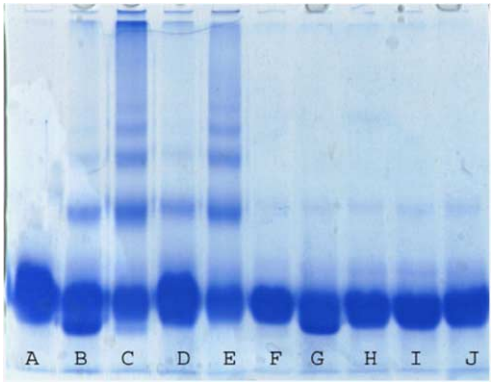


Figure 1. Protein crosslinking induced by unsaturated aldehydes. RNase or Me_2RNase (0.5 mM) was incubated with HNE, ONE, DDE, or DTE (5 mM) for 48 h in 50 mM pH 7.4 sodium phosphate buffer at 25 °C. Lane A: RNase control; B: RNase + HNE; C: RNase + ONE; D: RNase + DDE; E: RNase + DTE; F: Me_2RNase control; G: Me_2RNase + HNE; H: Me_2RNase + ONE; I: Me_2RNase + DDE; J: Me_2RNase + DTE.

The failure of the above one-step or one-pot procedures led us to consider various multi-step methods. The formation of α,β-unsaturated esters from saturated aldehydes or ketones and alkoxyacetylenes in the presence of a Lewis acid catalyst is well known.[13] It was reported that arylpolyene aldehydes could be converted into homologated esters using ethyl ethynyl ether in the presence of $BF_3{\cdot}O(C_2H_5)_2$.[14] Although there has been no report using this method to prepare aliphatic polyene α,β-unsaturated esters, we found that this protocol (Scheme 2) permitted the conversion of **1** to **3** quickly and cleanly at −30 to −20 °C. The reaction proceeded with greater than 95% *E* stereoselectivity (by NMR). Compound **3** was reduced by DIBAL in CH_2Cl_2 quantitatively to give **4**, which was converted into the known **2**[15] with MnO_2 in CH_2Cl_2 in high yield.[16]

Our research has demonstrated that the lipid peroxidation products HNE, ONE, and DDE, can readily modify and crosslink proteins.[7,17] A long term goal of this work is to understand which other types of biologically or environmentally relevant molecules might also have this activity. Data shown in Figure 1 indicates that DTE is a relatively potent protein crosslinking agent; not as potent as ONE, but more potent than DDE and HNE. The results suggest that extension of the conjugated C=C bond system increases reactivity toward nucleophilic protein side-chains. The finding that conversion of the accessible lysine primary ε-amino groups in RNase to tertiary dimethylamino groups (methylated RNase, prepared as described[17b]) results in complete abrogation of crosslinking, demonstrates that the lysine ε-amino group is an obligatory component of the crosslinks generated from these unsaturated aldehydes.

In conclusion, an efficient method for synthesizing the natural product 2*E*,4*E*,6*E*-dodecatrienal (DTE) was presented, and this method should be able to be extended to prepare other aliphatic polyene aldehydes. DTE was shown to be capable of protein crosslinking and is expected to react readily with DNA nucleobases.

Acknowledgements

This work was supported in part by National Institutes of Health grant HL53315.

References and notes

1. El-Shattory, Y.; Farag, R. S.; Aly, S. M.; Afifi, S. M. *Grasas Aceites* **2000**, *51*, 325–331.
2. Fadel, H. M.; Soliman, M. M. A. *Grasas Aceites* **1989**, *40*, 345–350.
3. Soliman, M. A.; El-Sawy, A. A.; Fadel, H. M.; Osman, F. *J. Agric. Food Chem.* **1985**, *33*, 523–528.
4. Nappez, C.; Battu, S.; Beneytout, J. L. *Cancer Lett.* **1996**, *99*, 115–119.
5. Loureiro, A. P.; de Arruda Campos, I. P.; Gomes, O. F.; di Mascio, P.; Medeiros, M. H. *Chem. Res. Toxicol.* **2004**, *17*, 641–649.
6. Beppu, M.; Murakami, K.; Kikugawa, K. *Chem. Pharm. Bull.* **1986**, *34*, 781–788.
7. Zhang, W.-H.; Liu, J.; Xu, G.; Yuan, Q.; Sayre, L. M. *Chem. Res. Toxicol.* **2003**, *16*, 512–523.
8. Sayre, L. M.; Yuan, Q. *Abstract of Papers*, 226th ACS National Meeting, New York, Sept 7–11, 2003, TOXI-086.
9. (a) Diaz, M. C.; Herranz, M. A.; Illescas, B. M.; Martin, N.; Godbert, N.; Bryce, M. R.; Luo, C.; Swartz, A.; Anderson, G.; Guldi, D. M. *J. Org. Chem.* **2003**, *68*, 7711–7721; (b) Sonoda, Y.; Nakao, Y. *J. Chem. Soc., Perkin Trans. 1* **1993**, 1147–1151; (c) Dann, O.; Wolff, H. P.; Schlee, R.; Ruff, J. *Liebigs Ann. Chem.* **1986**, 2164–2178; (d) Olstein, R.; Stephenson, E. F. M. *Aust. J. Chem.* **1979**, *32*, 681–686.
10. Maeta, H.; Suzuki, K. *Tetrahedron Lett.* **1993**, *34*, 341–344.
11. Bellassoued, M.; Majidi, A. *J. Org. Chem.* **1993**, *58*, 2517–2522.
12. Blank, I.; Lin, J.; Vera, F. A.; Welti, D. H.; Fay, L. B. *J. Agric. Food Chem.* **2001**, *49*, 2959–2965.
13. Oblin, M.; Pons, J.-M.; Parrain, J.-L.; Rajzmann, M. *Chem. Commun.* **1998**, 1619–1620.

14. Makin, S. M.; Mikerin, I. E.; Shavrygina, O. A.; Ermakova, G. A.; Arshava, B. M. *Zh. Org. Khim.* **1984**, *20*, 2317–2323.
15. Compound **2** was reported as a by-product: Nieuwenhuis, S. A. M.; Vertegaal, L. B. J.; de Zoete, M. C.; van der Gen, A. *Tetrahedron* **1994**, *50*, 13207–13230.
16. Preparation of **3**: to a solution of DDE (0.15 g, 1.0 mmol) in ether (20 mL) under argon cooled to −30 °C was added $BF_3{\cdot}O(C_2H_5)_2$ (0.15 mL, 1.2 mmol) in 10 mL of ether. After 30 min, with stirring at −30 to −25 °C, ethoxyacetylene (0.28 mL of a 40% solution in hexanes, 1.2 mmol) in 10 mL of ether was added slowly. After 1 h, saturated aqueous sodium carbonate solution was added, the ether layer was separated, and the aqueous layer was extracted with ether. The combined ether extract was dried (Na_2SO_4) and evaporated to give a yellow oil (0.16 g, 90%), which was used in the next step without further purification. ^{1}H NMR ($CDCl_3$): δ 7.28 (dd, 1H, J = 11.2, 15.2 Hz), 6.53 (dd, 1H, J = 11.2, 15.2 Hz), 6.09–6.25 (2m, 2H), 5.91 (m, 1H), 5.86 (d, 1H, J = 15.2 Hz), 4.20 (q, 2H, J = 7.1 Hz), 2.12 (q, 2H, J = 6.8 Hz), 1.25–1.48 (6H), 1.29 (t, 3H, J = 7.1 Hz), 0.89 (t, 3H, J = 6.8 Hz). Preparation of **4**: to a solution of **3** (0.16 g, 0.6 mmol) in 20 mL of CH_2Cl_2 under argon at −78 °C, was slowly added DIBAL (0.8 mL of 1.5 M in toluene, 1.2 mmol). The resulting mixture was stirred at −78 °C for 2.5 h, warmed to room temperature, and quenched by addition of 5 mL of water. The solid was filtered off, and the filtrate was extracted with CH_2Cl_2. The combined organic layer was dried (Na_2SO_4) and evaporated to give a light yellow oil (0.13 g, 99%), which was used directly in the next step. Preparation of **2**: to a solution of **4** (0.13 g, 0.6 mmol) in 20 mL of CH_2Cl_2 was added MnO_2 (0.6 g), and the mixture was stirred at room temperature for 72 h. The solid was filtered off, the filtrate was evaporated, and the crude product was purified by silica gel flash chromatography using hexanes–EtOAc (20:1, v/v) as eluant to give **2** (0.12 g, 99%). ^{1}H NMR ($CDCl_3$): δ 9.51 (d, 1H, J = 8.0 Hz), 7.09 (dd, 1H, J = 11.0, 15.2 Hz), 6.62 (dd, 1H, J = 10.0, 14.8 Hz), 6.31 (dd, 1H, J = 11.0, 14.8 Hz), 5.95–6.23 (3m, 3H), 2.10 (q, 2H, J = 6.9 Hz), 1.23–1.45 (6H), 0.87 (t, 3H, J = 6.7 Hz); ^{13}C NMR (APT, $CDCl_3$): δ 193.6 (−), 152.5 (−), 143.4 (−), 142.8 (−), 130.7 (−), 129.8 (−), 127.8 (−), 33.1 (+), 31.4 (+), 28.6 (+), 22.5 (+), 14.1 (−). HRMS (FAB) calcd for $C_{12}H_{17}O$ (M^+−H), 177.1279, found 177.1279.
17. (a) Xu, G.; Liu, Y.; Sayre, L. M. *J. Org. Chem.* **1999**, *64*, 5732–5745; (b) Xu, G.; Liu, Y.; Kansal, M. M.; Sayre, L. M. *Chem. Res. Toxicol.* **1999**, *12*, 855–861.