

Synthesis of deuterated 5-*n*-alkylresorcinols

Kirsti Parikka and Kristiina Wähälä*

Four alternative strategies for the preparation of deuterium poly-labelled 5-*n*-alkylresorcinols are explored. Ring-labelled $^2\text{H}_3$ -alkylresorcinols synthesized by acidic H/D exchange are stable under electrospray ionization MS conditions but scrambling occurs in electron bombardment ionization MS. Side chain-labelled $^2\text{H}_4$ -derivatives prepared by two different total synthesis approaches are contaminated by isotopologues with varying number of deuterium labels due to H/D redistribution and exchange during D_2 gas deuteration. The derivative carrying an ω - $^2\text{H}_3$ label is isotopically pure and completely stable under all relevant analytical conditions encountered in quantitation work.

Keywords: 5-*n*-alkylresorcinols; phenolic lipids; deuterium labelling; stable isotope

Introduction

5-Alk(en)ylresorcinols are non-isoprenoid phenolic lipids that carry an *n*-alkyl or alkenyl side chain at C-5 of the resorcinol ring. They are present in various families of plants (e.g. Gramineae and Anacardiaceae) and in some families of bacteria.¹ The use of 5-*n*-alkylresorcinols **1** (Figure 1) as biomarkers of whole grain intake and of a healthy diet is of great potential due to their presence in significant amounts in whole grain rye and wheat containing foods (from 300 mg/kg in whole wheat to 3000 mg/kg in whole rye).² 5-Alkylresorcinols have various biological effects including antioxidant activity,³ antimutagenic activity,⁴ antibacterial properties⁵ and inhibition of enzymes,⁶ for example.

The preparation of an isotopically labelled long chain 5-*n*-alkylresorcinol has been described only twice. ^{14}C -heneicosylresorcinol (C_{21}) was synthesized for a metabolism study⁷ and ^{13}C -dodecylresorcinol (C_{12}) was obtained by biosynthesis starting from a ^{13}C -labelled substrate.⁸ Two studies concern the labelling of short alkyl chain (C_5 , C_3) analogues which are substructures of tetrahydrocannabinols and their metabolites.⁹ The quantitative and qualitative screening of 5-*n*-alkylresorcinols from biological fluids has been performed using an even-chain analogue of alkylresorcinols as an internal standard,^{2,10} even though such even-chain alkylresorcinols have been detected from grains.¹¹ Deuterium-labelled 5-*n*-alkylresorcinol analogues with varying alkyl chain lengths would give a possibility to account more reliably for the losses during sample preparation in various quantitative measurement procedures.

We present here the synthesis of $^2\text{H}_3$ - or $^2\text{H}_4$ -labelled alkylresorcinols, aiming at single isotopomers of high isotopic purity and chemically stable labels. We have investigated different labelling strategies including acid-catalysed hydrogen/deuterium exchange (Scheme 1) or total synthesis (Schemes 2, 4, 5 and 6) where the labels are introduced into different parts of an alkylresorcinol structure.

Results and discussion

Acid-catalysed exchange experiments gave the $^2\text{H}_3$ -alkylresorcinols **2a–d** in good yields and isotopic purity (Scheme 1). We

found that microwave (MW)-assisted reactions provide an efficient entry to these products, using DCI or CF_3COOD and short reaction times. The mass spectra of the products **2** were run using electrospray ionization (ESI) due to scrambling problems¹² when electron bombardment ionization (EI) was used. These compounds, with a 3 amu increase in M^+ compared with the unlabelled analogue (Figure 2) to avoid peak overlap, should be useful as standards in LC-MS based analytical studies for example. High-performance liquid chromatography (HPLC) has been used in the analysis of 5-*n*-alkylresorcinols, but the validation procedures appear to be incomplete.^{2a}

To avoid scrambling problems and ensure the stability of the labels in the various analytical and pretreatment operations that sample preparation may require, for example, incubating under acidic conditions or derivatization, we next studied the introduction of four deuterium labels into the alkyl chain at the benzylic (α) position and the β site (Scheme 2). The four D atoms were introduced in a stepwise manner, involving the use of C-1 deuterated aldehyde (ArCDO) and C-1 dideuterated primary alcohol (RCD_2OH) starting materials and catalytic C=C reduction using D_2 gas. The $^2\text{H}_2$ -species **7** was homogenous by MS and NMR, but the product of the reduction ($^2\text{H}_4$ -nonadecylresorcinol dimethyl ether **8**) was contaminated by some $^2\text{H}_3$ -nonadecylresorcinol (up to 30%) and by a lesser amount of $^2\text{H}_2$ -nonadecylresorcinol (up to 10%) according to the mass spectra (EI, ESI). Thus, it appeared that D/H scrambling occurred in the C=C reduction. Earlier, the exchange and redistribution of hydrogen and deuterium have been observed when simple alkenes were deuterogenated over a nickel catalyst giving a mixture of alkanes with varying deuterium content, with the $^2\text{H}_4$ -derivative as the main product.¹³

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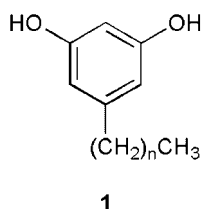
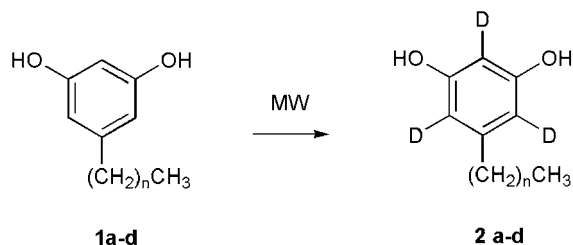


Figure 1. 5-*n*-alkylresorcinols present in whole grain products. *n* = 14/16/18/20/22.



Scheme 1

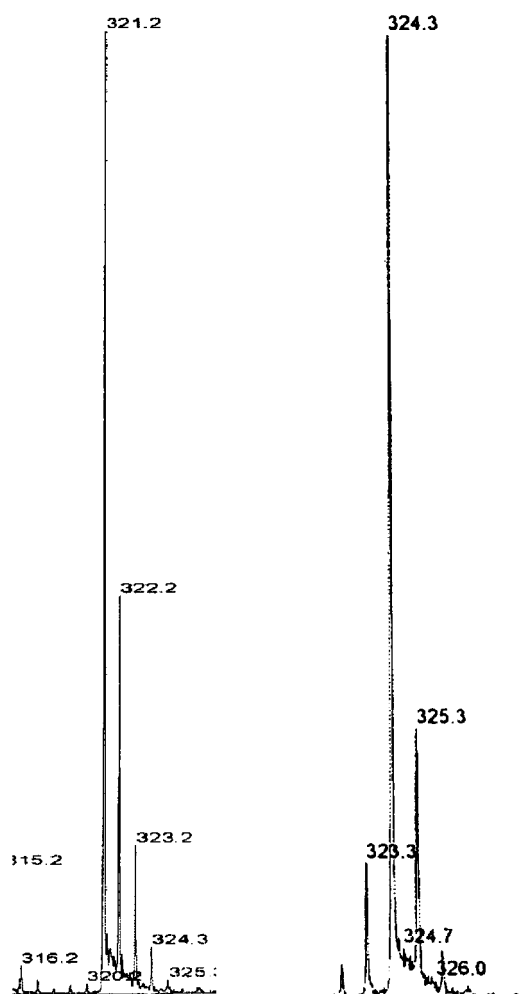


Figure 2. Molecular ion region in the mass spectra (M+H, ESI) of pentadecylresorcinol **1a** and D₃-pentadecylresorcinol **2a**.

Another alkene reduction strategy was investigated (Scheme 3). The deuteration of linoleic acid **10** (over Pd/C or Pd/BaSO₄), a potential starting material for alkylresorcinols, did not furnish the ²H₄-stearic acid **11** cleanly but gave instead a

mixture of ²H₀ up to ²H₁₅ isotopologues, ²H₄- and ²H₅-derivatives being the main products according to MS. There were no differences between the products of the two catalysts. Apparently, linoleic acid with the methylene skipped diene structure is much more prone to scrambling and redistribution reaction than the styrene derivative **7**.

Two variations of another strategy based on the deuteration of an alkyne to alkane were studied next (Schemes 4 and 5). However, D/H scrambling also occurred in the reduction of the acetylenic starting materials by D₂ gas. Deuterium atoms were introduced not only to the acetylenic carbons but also elsewhere in the alkyl chain, resulting in a mixture of isotopologous products (of **13** and subsequent synthetic intermediates **14–16**) according to MS and NMR. In the final alkylresorcinol derivative **17**, the ²H₄ species was the main product (ca. 29%) but ²H₂-, ²H₃- and at least ²H₅-²H₇-derivatives were present (approximately 11, 20, 20, 12 and 7%, respectively). In the case of **20** the distribution of the products was different and the heavier species were more abundant compared with **17**. The ²H₃-, ²H₄-, ²H₅- and ²H₆-derivatives were the main constituents in **20** (18, 20, 20 and 19%, respectively). The H/D exchange and redistribution have not been reported to occur to a major degree when alkynes are reduced selectively to alkenes,¹⁴ suggesting that the scrambling in our case occurs at the subsequent alkene reduction stage. However in our experiments, there was a difference between the products of the deuteration of either alkenes or alkynes. In contrast with the product **8** (and **9**) with only ²H₂- and ²H₃-derivatives present in addition to ²H₄, in the products **17** (and **13–16**) and **20** there were clearly significant amounts of ²H₅- and ²H₆-derivatives in the mixture. It seems that controlling the heterogeneous catalytic D₂ reduction of any olefinic or acetylenic starting material is problematic and specifically deuterated alkylresorcinols, or any long alkyl chain species for that matter, cannot be synthesized in this way.

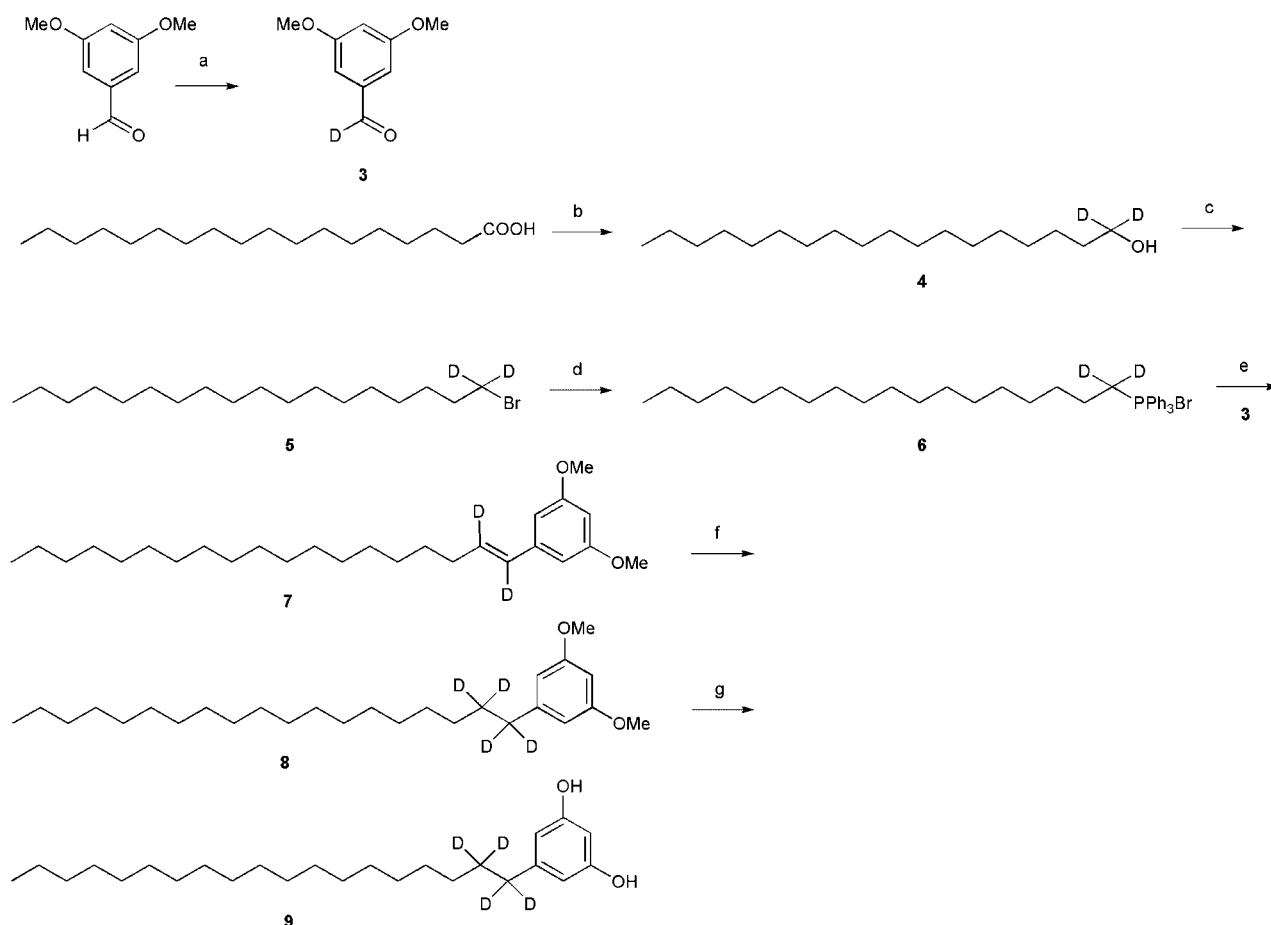
Finally, placing D atoms at the remote end of the alkyl chain in completely unactivating surroundings makes available derivatives (**25**, Scheme 6) that are not vulnerable to any kind of D/H exchanges whatsoever either in solution or in the mass spectrometer. A 5-(*ω*-methoxycarbonylalkyl)resorcinol dimethyl ether **21**[†] may be converted in four steps to *ω*-²H₃-alkylresorcinol with an isotopic purity in excess of 95% (Scheme 3). In the first step, two D atoms are introduced by LiAlD₄ reduction of the ester. The derived tosylate is very conveniently reduced by NaBD₄ in dimethyl sulphoxide (DMSO) in 8 min under MW irradiation (cf. the previously reported 2 h required for the preparation of alkanes with conventional heating¹⁵).

Experimental

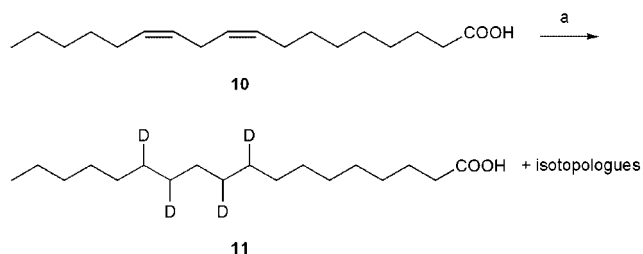
General

MW-assisted acid-catalysed deuteration were performed in a Teflon tube with a thread stopper in a domestic MW oven. The reaction with NaBD₄ was performed in a CEM Discover[®] system. DCI (37% in D₂O), LiAlD₄ (99% D) and NaBD₄ (99% D) were purchased from Aldrich. CF₃COOD was prepared according to the literature.¹⁶ Deuterated 3,5-dimethoxybenzaldehyde **3** was prepared as described in the literature.¹⁷ The NMR spectra of the synthesized compounds were obtained with a Varian 200 or 300 MHz spectrometer using tetramethylsilane as an internal standard. Mass spectra

[†]Parikka K and Wähälä K, unpublished work.



Scheme 2



Scheme 3

were obtained with a JEOL JMS SX102 mass spectrometer operating at 70 eV (EI), Varian Saturn 2000 instrument (GC-MS, EI), or with a Mariner ESI TOF (ESI) mass spectrometer. Flash chromatography was performed with silica gel 60.

[2,4,6-²H₃]-5-Pentadecylresorcinol 2a

5-Pentadecylresorcinol (0.1 g, 0.3 mmol) and DCI (2 ml) were MW irradiated in a Teflon tube, stopper slightly open, for 3 min. The procedure was then repeated two times. Each time the tube was allowed to cool down and 1.5 ml of DCI was added to replace the acid partly evaporated into the oven (CAUTION). The mixture was poured into water (50 ml) and the product extracted with EtOAc (3 × 15 ml). Recrystallization from cyclohexane afforded [2,4,6-²H₃]-5-pentadecylresorcinol (0.091 g, 91%), m.p. 89°C. Alter-

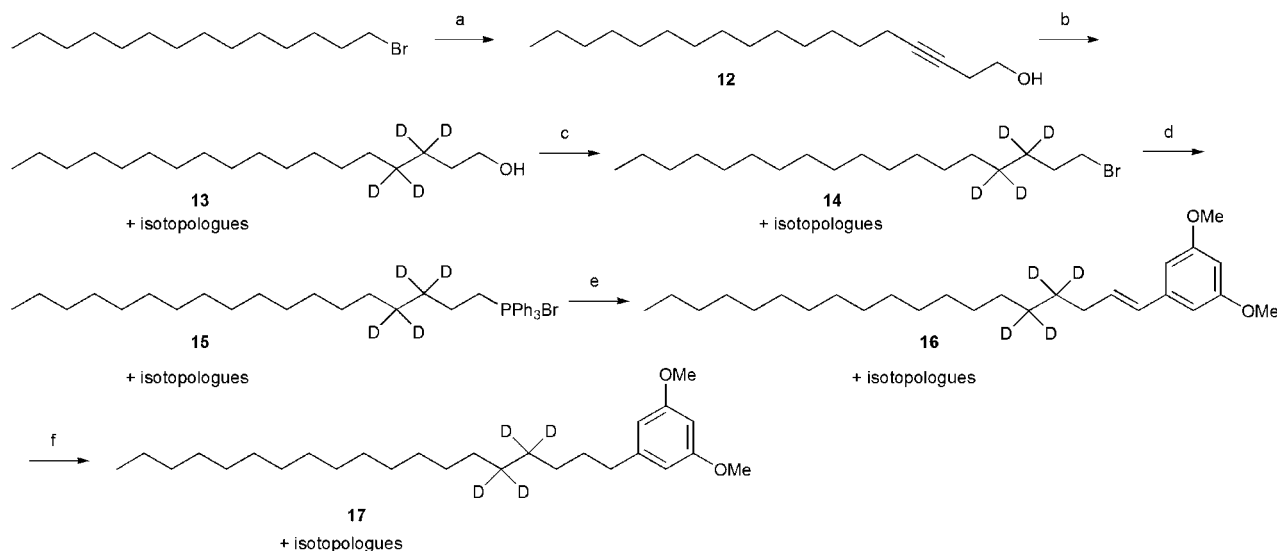
natively, 5-pentadecylresorcinol (0.07 g, 0.2 mmol) and freshly prepared CF₃COOD (2.5 ml) were MW irradiated in a similar Teflon tube for 5 min. The procedure was then repeated two times. Each time the tube was allowed to cool down and 2.5 ml of CF₃COOD was added to replace the acid evaporated into the oven (CAUTION). The mixture was poured into water (50 ml) and the product extracted with EtOAc (3 × 15 ml). Recrystallization from cyclohexane gave [2,4,6-²H₃]-5-pentadecylresorcinol (0.062 g, 89%), m.p. 89°C. ¹H NMR (200 MHz, D₆-acetone) δ 0.86–0.96 (m, 3H, CH₃), 1.20–1.35 (m, 14H, CH₂), 1.54–1.58 (m, 2H, CH₂), 2.45 (t, 2H, *J* = 7.7 Hz, CH₂Ar), 8.04 (br s, 2H, OH); MS (ESI) *m/z* (%) 323 (10), 324 (100) [M+H]⁺, 325 (20).

[2,4,6-²H₃]-5-Heptadecylresorcinol 2b

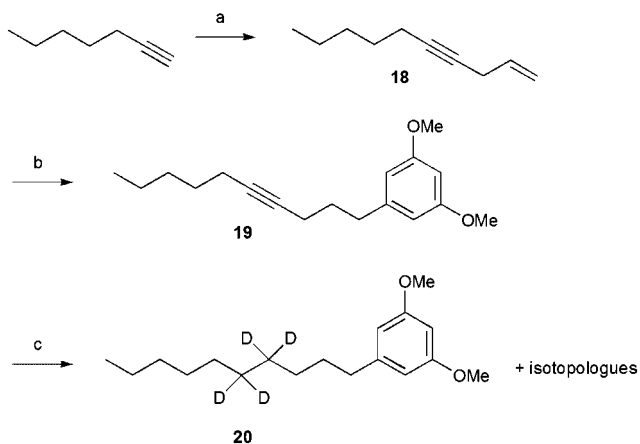
The reaction was performed as described for 5-pentadecylresorcinol. Recrystallization from cyclohexane gave [2,4,6-²H₃]-5-heptadecylresorcinol (85%), m.p. 91°C. ¹H NMR (200 MHz, D₆-acetone) δ 0.86–0.96 (m, 3H, CH₃), 1.20–1.35 (m, 18H, CH₂), 1.54–1.58 (m, 2H, CH₂), 2.45 (t, 2H, *J* = 7.7 Hz, CH₂Ar), 8.00 (br s, 2H, OH); MS (ESI) *m/z* (%) 351 (12), 352 (100) [M+H]⁺, 353 (30).

[2,4,6-²H₃]-Nonadecylresorcinol 2c

The reaction was performed as described for 5-pentadecylresorcinol. Recrystallization from cyclohexane gave [2,4,6-²H₃]-5-nonadecylresorcinol (87%), m.p. 92°C. ¹H NMR (200 MHz, D₆-acetone) δ 0.86–0.96 (m, 3H, CH₃), 1.20–1.35 (m, 22H, CH₂),



Scheme 4



Scheme 5

1.53–1.58 (m, 2H, CH₂), 2.45 (t, 2H, *J* = 7.7 Hz, CH₂Ar), 8.00 (br s, 2H, OH); MS (ESI) *m/z* (%) 379 (10), 380 (100) [M+H]⁺, 381 (40).

[2,4,6-²H₃]-5-Heneicosylresorcinol 2d

The reaction was performed as described for 5-pentadecylresorcinol. Recrystallization from cyclohexane gave [2,4,6-²H₃]-5-heneicosylresorcinol (90%), m.p. 97°C. ¹H NMR (200 MHz, D₆-acetone) δ 0.86–0.96 (m, 3H, CH₃), 1.20–1.35 (m, 26H, CH₂), 1.53–1.58 (m, 2H, CH₂), 2.45 (t, 2H, *J* = 7.7 Hz, CH₂Ar), 8.00 (br s, 2H, OH); MS (ESI) *m/z* (%) 407 (12), 408 (100) [M+H]⁺, 409 (40).

[1,1-²H₂]-1-Octadecanol 4

Octadecanoic acid (0.7 g, 2.3 mmol) and LiAlD₄ (0.37 g, 8.8 mmol) were refluxed in dry THF overnight. Water was added and the product extracted with EtOAc (3 × 20 ml). Combined extracts were washed with brine and dried over Na₂SO₄. Evaporation gave 4 as a white powder which was used in the next step without purification (0.49 g, 72%). ¹H NMR (200 MHz, CDCl₃) δ 0.85–0.91 (m, 3H, CH₃), 1.15–1.36 (m, 30H, CH₂), 1.52–1.58 (m, 2H, CH₂); MS (EI, 70 eV) *m/z* (%) 97 (100), 111 (70), 125 (30), 140 (20), 226 (15), 254 (30) [M–H₂O]⁺.

[1,1-²H₂]-1-Bromooctadecane 5

Concentrated HBr (45 ml), H₂SO₄ (3 ml) and 1,1-D₂-1-octadecanol 4 (0.45 g, 1.7 mmol) were refluxed for 4 h. Water was added and the product extracted with EtOAc (3 × 20 ml). The combined extracts were washed with saturated NaHCO₃. Evaporation gave 5 as a pale brown powder, which was used in the next step without purification (0.44 g, 79%). ¹H NMR (200 MHz, CDCl₃) δ 0.84–0.91 (m, 3H, CH₃), 1.15–1.36 (m, 30H, CH₂), 1.80–1.88 (m, 2H, CH₂); MS (EI, 70 eV) *m/z* (%) 71 (100), 85 (75), 137 (97), 139 (90), 151 (30), 153 (30), 255 (15), 334 (5), 336 (5).

[1,1-²H₂]-Octadecyltriphenylphosphonium bromide 6

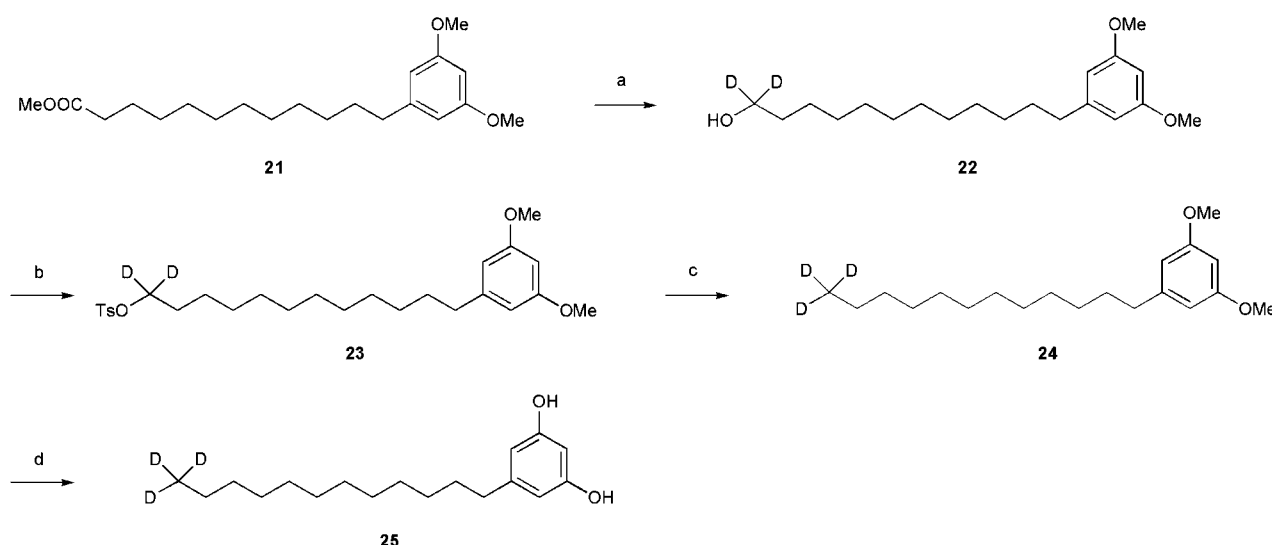
[1,1-²H₂]-1-Bromooctadecane 5 (0.43 g, 1.3 mmol) and PPh₃ (0.4 g, 1.5 mmol) were refluxed in dry toluene (10 ml) under argon for 22 h at 130°C. The solution was allowed to cool down and evaporated. The product 6 was crystallized from Et₂O as a white powder (0.64 g, 84%), m.p. 96°C. ¹H NMR (200 MHz, CDCl₃) δ 0.80–0.85 (m, 3H, CH₃), 1.17–1.35 (m, 30H, CH₂), 1.50–1.60 (m, 2H, CH₂), 7.65–7.99 (m, 15H, ArH).

[1,2-²H₂]-1-(3,5-Dimethoxyphenyl)-1-nonadecene 7

The phosphonium salt 6 (0.2 g, 0.34 mmol) was stirred with *n*-BuLi (0.43 ml, 1.2 M solution in hexane) in THF (10 ml) for 10 min at 0°C. ²H₁-3,5-Dimethoxybenzaldehyde 3 (0.056 g, 0.34 mmol) was added and the solution refluxed overnight. Water was added and the product extracted with Et₂O (3 × 20 ml). Combined extracts were washed with brine and dried over Na₂SO₄. The product was purified by flash chromatography eluting with hexane/CH₂Cl₂ 2/3 to give a colourless oil (0.092 g, 67%), a mixture of *E/Z* isomers (*E/Z* c.a. 60/40 according to GC-MS and ¹H NMR). MS (EI, 70 eV) *m/z* (%) 152 (30), 153 (100), 179 (20), 403 (10), 404 (65) M⁺, 405 (20).

[1,1,2,2-²H₄]-1-(3,5-Dimethoxyphenyl)nonadecane 8

[1,2-²H₂]-1-(3,5-Dimethoxyphenyl)-1-nonadecene 7 (0.025 g, 0.06 mmol) was stirred in CH₂Cl₂ containing 0.005 g of Pd/C (10% w/w) and connected to a balloon containing deuterium gas. The mixture was stirred overnight at room temperature.



Scheme 6

Filtration of the reaction mixture through Celite[®] gave **8** as a mixture of isotopologues (²H₂, 10%, ²H₃, 30%, ²H₄, 60%, 0.022 g, 90%). ¹H NMR (200 MHz, CDCl₃) δ 0.85–0.91 (m, 3H, CH₃), 1.13–1.38 (m, 30H, CH₂), 3.78 (s, 6H, OMe), 6.29 (t, 1H, *J* = 2.2 Hz, H-4'), 6.34 (d, 2H, *J* = 2.2 Hz, H-2' and H-6'); MS (EI, 70 eV) *m/z* (%) 153 (70), 154 (100), 406 (10), 407 (25), 408 (50) M⁺, 409 (20).

[1,1,2,2-²H₄]-1-(3,5-Dihydroxyphenyl)nonadecane **9**

[1,1,2,2-²H₄]-1-(3,5-Dimethoxyphenyl)nonadecane **8** (0.02 g, 0.05 mmol) was stirred with BBr₃ (0.44 ml, 1 M solution in CH₂Cl₂) in CH₂Cl₂ (5 ml) under argon at 0°C until the reaction was complete according to TLC. Water was added and the product extracted with EtOAc (3 × 10 ml). The combined extracts were washed with saturated NaHCO₃ and dried over Na₂SO₄. Flash chromatography on silica gel eluting with MeOH/EtOAc/CH₂Cl₂ 1/2/7 gave **9** as a mixture of isotopologues (²H₂, 10%, ²H₃, 40%, ²H₄, 50%, 0.015 g, 79%). ¹H NMR (200 MHz, D₆-acetone) δ 0.86–0.96 (m, 3H, CH₃), 1.20–1.35 (m, 30H, CH₂), 6.18 (s, 3H, H-2', H-4' and H-6'), 8.14 (s, 2H, OH); MS (EI, 70 eV) *m/z* (%) 125 (65), 126 (100), 378 (7), 379 (25), 380 (37) M⁺, 381 (10).

Deuteration of linoleic acid

Linoleic acid (0.5 g, 1.8 mmol) was stirred in CH₂Cl₂ containing 0.05 g of Pd/C (10% w/w) and connected to a balloon containing deuterium gas. The mixture was stirred overnight at room temperature. Filtration of the reaction mixture through Celite[®] gave a mixture of deuterated stearic acids (²H₀–²H₁₅, ²H₄ and ²H₅ the main isotopologues) (0.51 g, 98%). A similar mixture (²H₀–²H₁₅) was obtained using Pd/BaSO₄ (0.1 g, 5% w/w) instead of Pd/C (0.51 g, 99%). ¹H NMR (200 MHz, CDCl₃) δ 0.85–0.91 (m, 3H, CH₃), 1.15–1.40 (m, c.a. 24H, CH₂), 1.60–1.67 (m, 2H, CH₂), 2.34 (t, 2H, *J* = 7.2 Hz, H-2); MS (EI, 70 eV) *m/z* (%) 97 (25), 98 (40), 99 (25), 115 (30), 116 (20), 129 (90), 130 (40), 131 (17), 284 (10), 285 (27), 286 (60), 287 (80), 288 (92), 289 (92), 290 (85), 291 (75), 292 (65), 293 (55), 294 (47), 295 (40), 296 (33), 297 (25), 298 (20), 299 (15).

3-Octadecyn-1-ol **12**

3-Butyn-1-ol (0.3 g, 0.0043 mmol), HMPA (2.3 g, 12.9 mmol, 2.3 ml) and *n*-BuLi (1.2 M in hexane, 7.2 ml) were stirred in THF

(10 ml) at 72°C for 1 h. 1-Bromotetradecane (0.6 g, 2.2 mmol) was added and the solution was stirred at room temperature overnight. The solution was acidified with 0.1 M HCl and extracted with Et₂O (3 × 30 ml). The combined extracts were dried over MgSO₄. Flash chromatography on silica gel eluting with hexane/acetone 1/1 gave **12** (0.41 g, 52%) as an amorphous solid. ¹H NMR (200 MHz, CDCl₃) δ 0.83–0.90 (m, 3H, CH₃), 1.12–1.38 (m, 22H, CH₂), 1.40–1.55 (m, 2H, CH₂), 2.10–2.18 (m, 2H, H-5), 2.37–2.46 (m, 2H, H-2), 3.66 (t, 2H, *J* = 6.2 Hz, H-1); ¹³C NMR (200 MHz, CDCl₃) δ 15.4, 20.0, 24.0, 24.5, 30.2, 30.3, 30.5, 30.6, 30.8, 31.0, 33.2, 62.7, 77.5, 84.1; MS (EI, 70 eV) *m/z* (%) 55 (100), 69 (97), 84 (100), 97 (100), 107 (73), 109 (67), 121 (50), 135 (30), 153 (30), 223 (20), 237 (10), 265 (8), 266 (15).

Deuterated octadecanol **13**

3-Octadecyn-1-ol **12** (0.14 g, 0.53 mmol) was stirred in CH₂Cl₂ containing 0.015 g of Pd/C (10% w/w) and connected to a balloon containing deuterium gas. The mixture was stirred overnight at room temperature. Filtration of the reaction mixture through Celite[®] gave a mixture of isotopologues (²H₂–²H₇, ²H₃–²H₅ as the main products, 0.13 g, 93%). ¹H NMR (200 MHz, CDCl₃) δ 0.84–0.90 (m, 3H, CH₃), 1.12–1.37 (m, ca. 26H, CH₂), 1.50–1.56 (m, 2H, CH₂), 3.62 (t, 2H, *J* = 6.6 Hz, H-1).

Deuterated bromooctadecane **14**

Concentrated HBr (15 ml), H₂SO₄ (1 ml) and **13** (0.092 g, 0.34 mmol) were refluxed for 4 h. Water was added and the product extracted with EtOAc (3 × 20 ml). The combined extracts were washed with saturated NaHCO₃. Evaporation gave **14** as a pale yellowish powder, which was used in the next step without purification (isotopologues ²H₂–²H₇, ²H₃–²H₅ as the main products, 0.087 g, 77%). ¹H NMR (200 MHz, CDCl₃) δ 0.85–0.91 (m, 3H, CH₃), 1.13–1.38 (m, ca. 26H, CH₂), 1.79–1.89 (m, 2H, CH₂), 3.40 (t, 2H, *J* = 6.6 Hz, H-1).

Deuterated octadecyltriphenylphosphonium bromide **15**

Deuterated bromooctadecane **14** (0.087 g, 0.26 mmol) and PPh₃ (0.074 g, 0.29 mmol) were refluxed in dry toluene (5 ml) under

argon for 20 h at 130°C. The solution was allowed to cool down and evaporated. The product was crystallized from Et₂O as a white powder (isotopologues ²H₂-²H₇, ²H₃-²H₅ as the main products, 0.11 g, 71%). ¹H NMR (200 MHz, CDCl₃) δ 0.82–0.87 (m, 3H, CH₃), 1.18–1.36 (m, ca. 26H, CH₂), 1.56–1.66 (m, 2H, CH₂), 3.66–3.86 (m, 2H, H-1), 7.54–7.88 (m, 15H, ArH).

Deuterated 1-(3,5-dimethoxyphenyl)-1-nonadecene 16

The phosphonium salt **15** (0.10 g, 0.17 mmol) was stirred with *n*-BuLi (0.15 ml, 1.2 M solution in hexane) in THF (10 ml) for 10 min under argon at 0°C. 3,5-Dimethoxybenzaldehyde (0.028 g, 0.17 mmol) was added and the solution refluxed overnight. Water was added and the product extracted with Et₂O (3 × 20 ml). Combined extracts were washed with brine and dried over Na₂SO₄. The product was purified by flash chromatography eluting with hexane/CH₂Cl₂ 2/3 to give a colourless oil (isotopologues ²H₂, 10%; ²H₃, 21%; ²H₄, 31%; ²H₅, 13%; ²H₆, 17%; ²H₇, 6%; 0.045 g, 65%). MS (EI, 70 eV) *m/z* (%) 151 (20), 152 (100), 153 (50), 154 (20), 179 (25), 404 (15), 405 (35), 406 (50), 407 (35), 408 (25), 409 (15).

Deuterated 1-(3,5-dimethoxyphenyl)nonadecane 17

The nonadecene **16** (0.01 g, 0.025 mmol) was hydrogenated by H₂ gas in CH₂Cl₂ containing 0.001 g of Pd/C (10% w/w) under atmospheric pressure and room temperature. Filtration of the reaction mixture through Celite® gave **17** (isotopologues ²H₂, 11%; ²H₃, 20%; ²H₄, 29%; ²H₅, 20%; ²H₆, 12%; ²H₇, 7%; 0.009 g, 89%). MS (EI, 70 eV) *m/z* (%) 151 (20), 152 (100), 153 (60), 154 (25), 406 (12), 407 (25), 408 (30), 409 (25), 410 (20), 411 (10).

1-Decen-4-yne 18

1-Heptyne (0.50 g, 0.52 mmol, 0.68 ml), *n*-BuLi (4.2 ml, 1.25 M solution in hexane) and THF (7 ml) were stirred at –72°C under argon for 1 h. 1-Bromopropene (0.63 g, 0.52 mmol, 0.44 ml) was added and the solution stirred at room temperature overnight. Water was added and the product extracted with hexane (2 × 30 ml). The combined extracts were dried over MgSO₄. Distillation under reduced pressure gave **18** as a colourless oil (0.3 g, 43%), b.p. 72–73°C/22 mmHg (Lit. 73–74°C/22 mmHg¹⁸). ¹H NMR (200 MHz, CDCl₃) δ 0.85–0.93 (m, 3H, CH₃), 1.31–1.55 (m, 6H, CH₂), 2.10–2.25 (m, 2H, H-6), 2.90–2.97 (m, 2H, H-3), 5.05–5.15 (m, 1H, H-1), 5.25–5.35 (m, 1H, H-1), 5.75–5.95 (m, 1H, H-2).

1-(3,5-dimethoxyphenyl)-4-decyne 19

1-Decen-4-yne **18** (0.05 g, 0.37 mmol), 9-BBN (1.5 ml, 0.5 M solution in THF) and dry THF (11 ml) were stirred under argon at room temperature for 2.5 h. NaOMe (0.046 g, 0.86 mmol), PdCl₂ (dppf) (0.016 g, 0.02 mmol) and 3,5-dimethoxyphenol trifluoromethanesulphonate (0.19 g, 0.66 mmol) were added and the mixture refluxed for 2 h. Water was added and the product extracted with Et₂O (3 × 20 ml). The combined extracts were washed with brine and dried over Na₂SO₄. The product was purified by flash chromatography eluting with CH₂Cl₂/hexane 2/1 to give a colourless oil (0.024 g, 24%). ¹H NMR (200 MHz, CDCl₃) δ 0.88–0.94 (m, 3H, CH₃), 1.34–1.57 (m, 6H, CH₂), 1.75–1.85 (m, 2H, CH₂), 2.14–2.20 (m, 4H, CH₂), 2.66 (t, 2H, *J* = 7.6 Hz, CH₂Ar), 3.78 (s, 6H, OCH₃), 6.30 (t, 1H, *J* = 2.2 Hz, H-4), 6.36 (d, 2H, *J* = 2.2 Hz, H-2 and H-6); ¹³C NMR (200 MHz, CDCl₃) δ 14.6, 18.9, 19.3, 22.8, 29.5, 31.1, 31.7, 35.7, 55.8, 80.2, 81.5, 98.4,

107.2, 144.9, 161.3; MS (EI, 70 eV) *m/z* (%) 152 (100), 191 (25), 203 (10), 217 (12), 259 (7), 274 (5) M⁺.

Deuteration of 19

1-(3,5-Dimethoxyphenyl)-4-decyne (**19**) (0.024 g, 0.087 mmol) was stirred in CH₂Cl₂ containing 0.003 g of Pd/C (10% w/w) and connected to a balloon containing deuterium gas. The mixture was stirred overnight at room temperature. Filtration of the reaction mixture through Celite® gave **20** (isotopologues ²H₃, 18%; ²H₄, 20%; ²H₅, 20%; ²H₆, 19% as the main products; 0.023 g, 95%). MS (EI, 70 eV) *m/z* (%) 151 (10), 152 (90), 153 (100), 154 (60), 155 (20), 281 (11), 282 (14), 283 (13), 284 (13), 285 (9), 286 (6).

[1,1-²H₂]-12-(3,5-Dimethoxyphenyl)dodecan-1-ol 22

Methyl 12-(3,5-dimethoxyphenyl)dodecanoate **21** (0.075 g, 0.21 mmol) and LiAlD₄ (0.040 g, 0.84 mmol) were refluxed in dry THF for 3 h. Water was added and the mixture was acidified with 0.1 M HCl. The product was extracted with EtOAc (3 × 10 ml). Combined extracts were washed with brine and dried over Na₂SO₄. Evaporation gave **22** as a white powder which was used in the next step without purification (0.070 g, 93%). ¹H NMR (300 MHz, CDCl₃) δ 1.26–1.35 (m, 16H, CH₂), 1.53–1.60 (m, 4H, CH₂), 2.54 (t, 2H, *J* = 7.6 Hz, CH₂Ar), 3.78 (s, 6H, OMe), 6.29 (t, 1H, *J* = 2.1 Hz, H-4'), 6.34 (d, 2H, *J* = 2.1 Hz, H-2' and H-6'); ¹³C NMR (300 MHz, CDCl₃) δ 25.7, 29.3, 29.4, 29.6, 31.3, 31.8, 32.6, 36.3, 55.2, 97.6, 106.5, 145.4, 160.7.

[1,1-²H₂]-12-(3,5-Dimethoxyphenyl)dodecyl toluenesulphonate 23

[1,1-²H₂]-12-(3,5-Dimethoxyphenyl)dodecan-1-ol **22** (0.06 g, 0.18 mmol), toluenesulphonyl chloride (0.041 g, 0.22 mmol) and Et₃N (0.04 ml) were stirred in CH₂Cl₂ (5 ml) at room temperature overnight. Water was added and the product extracted with CH₂Cl₂ (3 × 10 ml). Combined extracts were washed with brine and dried over Na₂SO₄. Flash chromatography on silica gel eluting with CH₂Cl₂ gave **23** (0.053 g, 62%). ¹H NMR (300 MHz, CDCl₃) δ 1.21–1.29 (m, 16H, CH₂), 1.53–1.64 (m, 4H, CH₂), 2.44 (s, 3H, CH₃), 2.54 (t, 2H, *J* = 7.6 Hz, CH₂Ar), 3.78 (s, 6H, OMe), 6.29 (t, 1H, *J* = 2.1 Hz, H-4'), 6.34 (d, 2H, *J* = 2.1 Hz, H-2' and H-6'), 7.34 (d, 2H, *J* = 8.0 Hz, ArH), 7.79 (d, 2H, *J* = 8.0 Hz, ArH); ¹³C NMR (300 MHz, CDCl₃) δ 21.6, 25.3, 28.6, 28.9, 29.4, 29.5, 31.3, 36.3, 55.2, 97.6, 106.5, 127.9, 129.8, 145.4, 160.7; MS (EI, 70 eV) *m/z* (%) 91 (45), 151 (45), 152 (100), 194 (10), 306 (10), 478 (20) M⁺, 479 (5).

[12,12,12-²H₃]-1-(3,5-Dimethoxyphenyl)dodecane 24

[1,1-²H₂]-12-(3,5-Dimethoxyphenyl)dodecyl toluenesulphonate **23** (0.050 g, 0.1 mmol) and NaBD₄ (0.009 g, 0.21 mmol) were stirred under MW irradiation (100 W, 85°C) for 8 min in DMSO (2 ml). Water was added and the product extracted with Et₂O (3 × 10 ml). The combined extracts were dried over Na₂SO₄. Flash chromatography on silica gel eluting with CH₂Cl₂ gave **24** (0.019 g, 63%). ¹H NMR (300 MHz, CDCl₃) δ 1.26–1.31 (m, 18H, CH₂), 1.53–1.64 (m, 2H, CH₂), 2.54 (t, 2H, *J* = 7.8 Hz, CH₂Ar), 3.78 (s, 6H, OMe), 6.30 (t, 1H, *J* = 2.1 Hz, H-4'), 6.35 (d, 2H, *J* = 2.1 Hz, H-2' and H-6'); ¹³C NMR (300 MHz, CDCl₃) δ 13.9, 22.4, 29.4, 29.5, 29.7, 31.3, 31.9, 36.3, 55.2, 97.6, 106.5, 145.5, 160.7; MS (EI, 70 eV) *m/z* (%) 121 (10), 151 (55), 152 (100), 165 (40), 194 (10), 308 (5), 309 (30) M⁺, 310 (5).

[12,12,12-²H₃]-1-(3,5-Dihydroxyphenyl)dodecane 25

[12,12,12-²H₃]-1-(3,5-Dimethoxyphenyl)dodecane **24** (0.016 g, 0.05 mmol) was stirred with BBr₃ (0.21 ml, 1 M solution in CH₂Cl₂) in CH₂Cl₂ (5 ml) under argon at 0°C until the reaction was complete according to TLC. Water was added and the product extracted with EtOAc (3 × 10 ml). The combined extracts were washed with saturated NaHCO₃ and dried over Na₂SO₄. Flash chromatography on silica gel eluting with MeOH/CH₂Cl₂ 1/9 gave **25** (0.012 g, 80%). ¹H NMR (300 MHz, D₆-acetone) δ 1.20–1.35 (m, 18H, CH₂), 1.54–1.58 (m, 2H, CH₂), 2.44 (t, 2H, *J* = 7.6 Hz, CH₂Ar), 6.18 (s, 3H, H-2', H-4' and H-6'), 7.96 (s, 2H, OH); ¹³C NMR (300 MHz, D₆-acetone) δ 13.7, 23.0, 29–30 (overlapping with acetone), 32.0, 32.5, 36.5, 100.9, 107.6, 145.7, 159.2; MS (EI, 70 eV): *m/z* (%) 88 (30), 123 (30), 124 (100), 138 (60), 149 (10), 280 (3), 281 (20) M⁺, 282 (5).

Conclusion

We obtained **2a–d** in approximately 90% isotopic purity and we suggest that these types of compounds would be practical in analytical studies that are based on ESI MS. The product **9** might be utilized as a standard since the M⁺ of the ²H₂-species does not overlap with any peak of the unlabelled analogue. Alkene deuterogenation strategies lead to extensive H/D scrambling along the alkyl chain. The investigation of different deuteration strategies proved that the deuteration method giving product **25** was the most reliable. The isotopic purity exceeds 95% for **25** and no H/D scrambling is expected under any conditions. Deuterated alkylresorcinols prepared by this synthesis route would constitute highly suitable standards for metabolic and analytical studies.

Acknowledgement

Jenny and Antti Wihuri Foundation, the Academy of Finland (205781) and the Graduate School of Bio-organic and Medicinal Chemistry are gratefully acknowledged for partial financial support.

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