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Synthesis of deuterated 5-*n*-alkylresorcinols

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Four alternative strategies for the preparation of deuterium poly-labelled 5-*n*-alkylresorcinols are explored. Ring-labelled $^2H_3$-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 $^2H_4$-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 deuterogenation. The derivative carrying an ω-$^2H_3$ 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 properties[5] 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 study[7] 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 $^2H_3$- or $^2H_4$-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 $^2H_3$-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 DCl or CF_3COOD and short reaction times. The mass spectra of the products **2** were run using electrospray ionization (ESI) due to scrambling problems[12] 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 $^2H_2$-species **7** was homogenous by MS and NMR, but the product of the reduction ($^2H_4$-nonadecylresorcinol dimethyl ether **8**) was contaminated by some $^2H_3$-nonadecylresorcinol (up to 30%) and by a lesser amount of $^2H_2$-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 $^2H_4$-derivative as the main product.[13]

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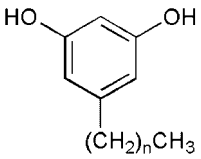

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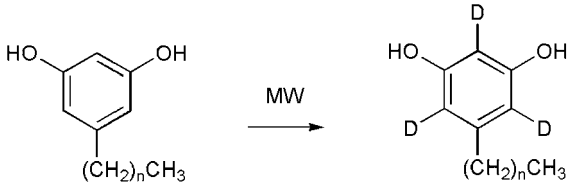
Figure 1. 5-*n*-alkylresorcinols present in whole grain products. n = 14/16/18/20/22.



1a-d **2 a-d**

Scheme 1

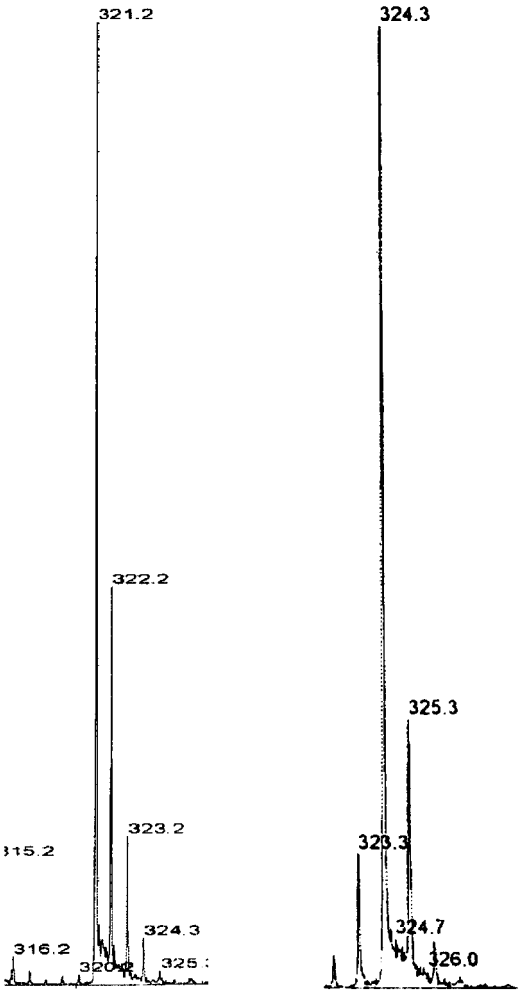


Figure 2. Molecular ion region in the mass spectra (M+H, ESI) of pentadecylresorcinol **1a** and D_3-pentadecylresorcinol **2a**.

Another alkene reduction strategy was investigated (Scheme 3). The deuterogenation of linoleic acid **10** (over Pd/C or Pd/$BaSO_4$), a potential starting material for alkylresorcinols, did not furnish the $^2H_4$-stearic acid **11** cleanly but gave instead a mixture of $^2H_0$ up to $^2H_{15}$ isotopologues, $^2H_4$- and $^2H_5$-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 deuterogenation 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 D_2 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 $^2H_4$ species was the main product (ca. 29%) but $^2H_2$-, $^2H_3$- and at least $^2H_5$-$^2H_7$-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 $^2H_3$-, $^2H_4$-, $^2H_5$- and $^2H_6$-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 deuterogenation of either alkenes or alkynes. In contrast with the product **8** (and **9**) with only $^2H_2$- and $^2H_3$-derivatives present in addition to $^2H_4$, in the products **17** (and **13–16**) and **20** there were clearly significant amounts of $^2H_5$- and $^2H_6$-derivatives in the mixture. It seems that controlling the heterogeneous catalytic D_2 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.

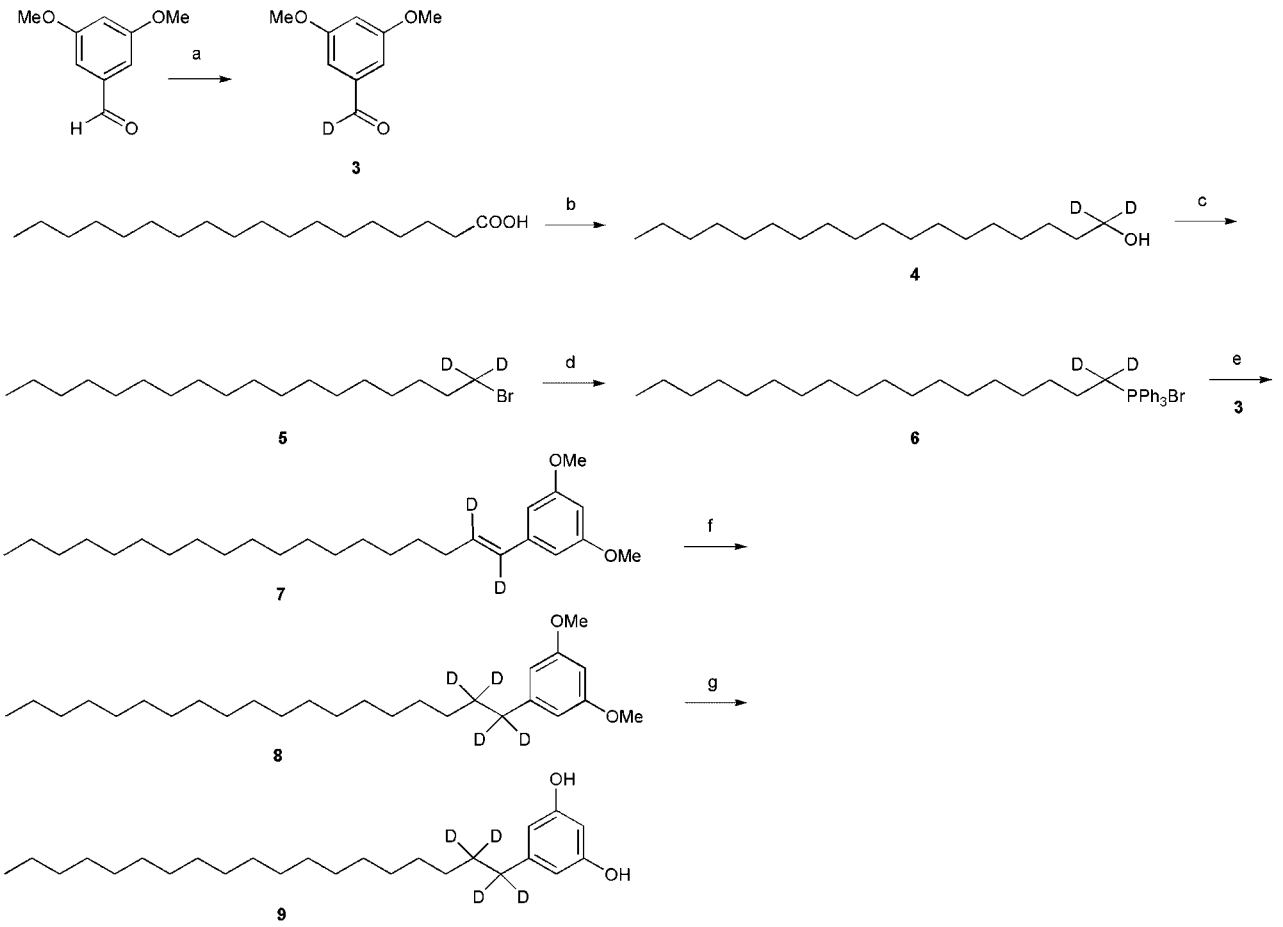
Finally, placing D atoms at the remote end of the alkyl chain in completely unactivating surroundings makes available derivatives (**25**, Scheme 6) that are not vulnerable to any kind of D/H exchanges whatsoever either in solution or in the mass spectrometer. A 5-(ω-methoxycarbonylalkyl)resorcinol dimethyl ether **21**[†] may be converted in four steps to ω-$^2H_3$-alkylresorcinol with an isotopic purity in excess of 95% (Scheme 3). In the first step, two D atoms are introduced by $LiAlD_4$ reduction of the ester. The derived tosylate is very conveniently reduced by $NaBD_4$ in dimethyl sulphoxide (DMSO) in 8 min under MW irradiation (cf. the previously reported 2 h required for the preparation of alkanes with conventional heating[15]).

Experimental

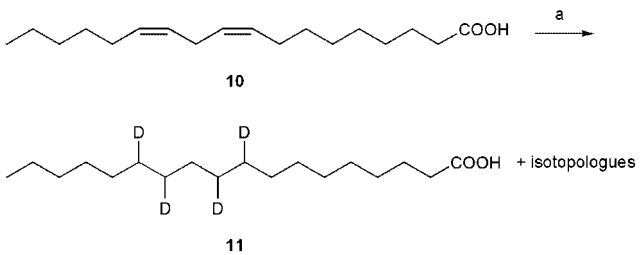
General

MW-assisted acid-catalysed deuterations were performed in a Teflon tube with a thread stopper in a domestic MW oven. The reaction with $NaBD_4$ was performed in a CEM Discover® system. DCl (37% in D_2O), $LiAlD_4$ (99% D) and $NaBD_4$ (99% D) were purchased from Aldrich. CF_3COOD 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-$^2H_3$]-5-Pentadecylresorcinol 2a

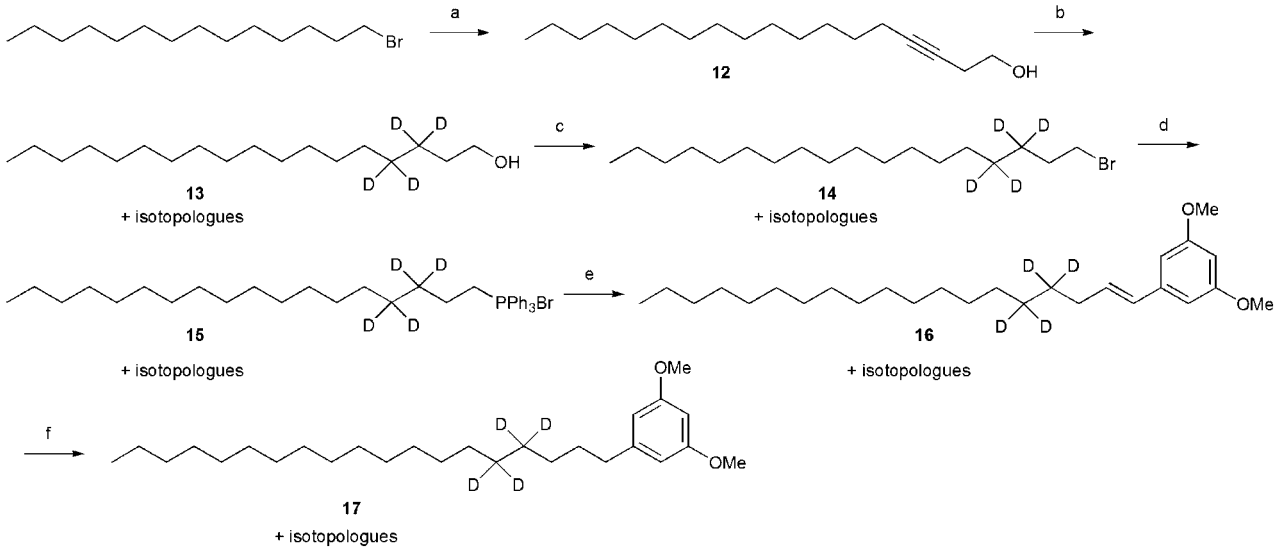
5-Pentadecylresorcinol (0.1 g, 0.3 mmol) and DCl (2 ml) were MW irradiated in a Teflon tube, stopper slightly open, for 3 min. The procedure was then repeated two times. Each time the tube was allowed to cool down and 1.5 ml of DCl was added to replace the acid partly evaporated into the oven (CAUTION). The mixture was poured into water (50 ml) and the product extracted with EtOAc (3 × 15 ml). Recrystallization from cyclohexane afforded [2,4,6-$^2H_3$]-5-pentadecylresorcinol (0.091 g, 91%), m.p. 89°C. Alternatively, 5-pentadecylresorcinol (0.07 g, 0.2 mmol) and freshly prepared CF_3COOD (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 CF_3COOD 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-$^2H_3$]-5-pentadecylresorcinol (0.062 g, 89%), m.p. 89°C. ^{1}H NMR (200 MHz, D_6-acetone) δ 0.86–0.96 (m, 3H, CH_3), 1.20–1.35 (m, 14H, CH_2), 1.54–1.58 (m, 2H, CH_2), 2.45 (t, 2H, $J = 7.7$ Hz, CH_2Ar), 8.04 (br s, 2H, OH); MS (ESI) *m/z* (%) 323 (10), 324 (100) $[M+H]^+$, 325 (20).

[2,4,6-$^2H_3$]-5-Heptadecylresorcinol 2b

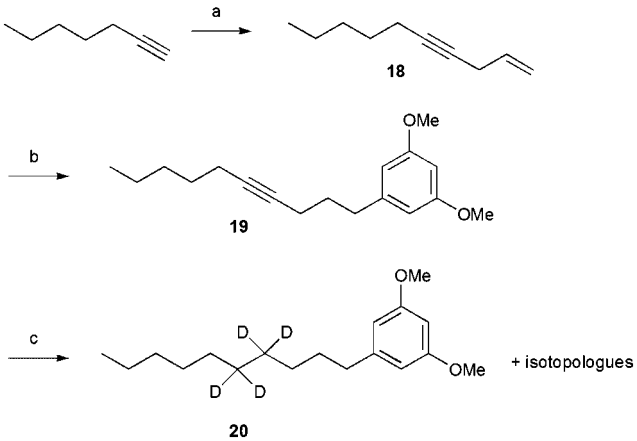
The reaction was performed as described for 5-pentadecylresorcinol. Recrystallization from cyclohexane gave [2,4,6-$^2H_3$]-5-heptadecylresorcinol (85%), m.p. 91°C. ^{1}H NMR (200 MHz, D_6-acetone) δ 0.86–0.96 (m, 3H, CH_3), 1.20–1.35 (m, 18H, CH_2), 1.54–1.58 (m, 2H, CH_2), 2.45 (t, 2H, $J = 7.7$ Hz, CH_2Ar), 8.00 (br s, 2H, OH); MS (ESI) *m/z* (%) 351 (12), 352 (100) $[M+H]^+$, 353 (30).

[2,4,6-$^2H_3$]Nonadecylresorcinol 2c

The reaction was performed as described for 5-pentadecylresorcinol. Recrystallization from cyclohexane gave [2,4,6-$^2H_3$] 5-nonadecylresorcinol (87%), m.p. 92°C. ^{1}H NMR (200 MHz, D_6-acetone) δ 0.86–0.96 (m, 3H, CH_3), 1.20–1.35 (m, 22H, CH_2),



Scheme 4



Scheme 5

1.53–1.58 (m, 2H, CH_2), 2.45 (t, 2H, $J = 7.7$ Hz, CH_2Ar), 8.00 (br s, 2H, OH); MS (ESI) *m/z* (%) 379 (10), 380 (100) $[M+H]^+$, 381 (40).

[2,4,6-$^2H_3$]-5-Heneicosylresorcinol 2d

The reaction was performed as described for 5-pentadecylresorcinol. Recrystallization from cyclohexane gave [2,4,6-$^2H_3$]-5-heneicosylresorcinol (90%), m.p. 97°C. ^1H NMR (200 MHz, D_6-acetone) δ 0.86–0.96 (m, 3H, CH_3), 1.20–1.35 (m, 26H, CH_2), 1.53–1.58 (m, 2H, CH_2), 2.45 (t, 2H, $J = 7.7$ Hz, CH_2Ar), 8.00 (br s, 2H, OH); MS (ESI) *m/z* (%) 407 (12), 408 (100) $[M+H]^+$, 409 (40).

[1,1-$^2H_2$]-1-Octadecanol 4

Octadecanoic acid (0.7 g, 2.3 mmol) and $LiAlD_4$ (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 Na_2SO_4. Evaporation gave **4** as a white powder which was used in the next step without purification (0.49 g, 72%). ^1H NMR (200 MHz, $CDCl_3$) δ 0.85–0.91 (m, 3H, CH_3), 1.15–1.36 (m, 30H, CH_2), 1.52–1.58 (m, 2H, CH_2); MS (EI, 70 eV) *m/z* (%) 97 (100), 111 (70), 125 (30), 140 (20), 226 (15), 254 (30) $[M-H_2O]^+$.

[1,1-$^2H_2$]-1-Bromooctadecane 5

Concentrated HBr (45 ml), H_2SO_4 (3 ml) and 1,1-D_2-1-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 $NaHCO_3$. 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, $CDCl_3$) δ 0.84–0.91 (m, 3H, CH_3), 1.15–1.36 (m, 30H, CH_2), 1.80-1.88 (m, 2H, CH_2); 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-$^2H_2$]Octadecyltriphenylphosphonium bromide 6

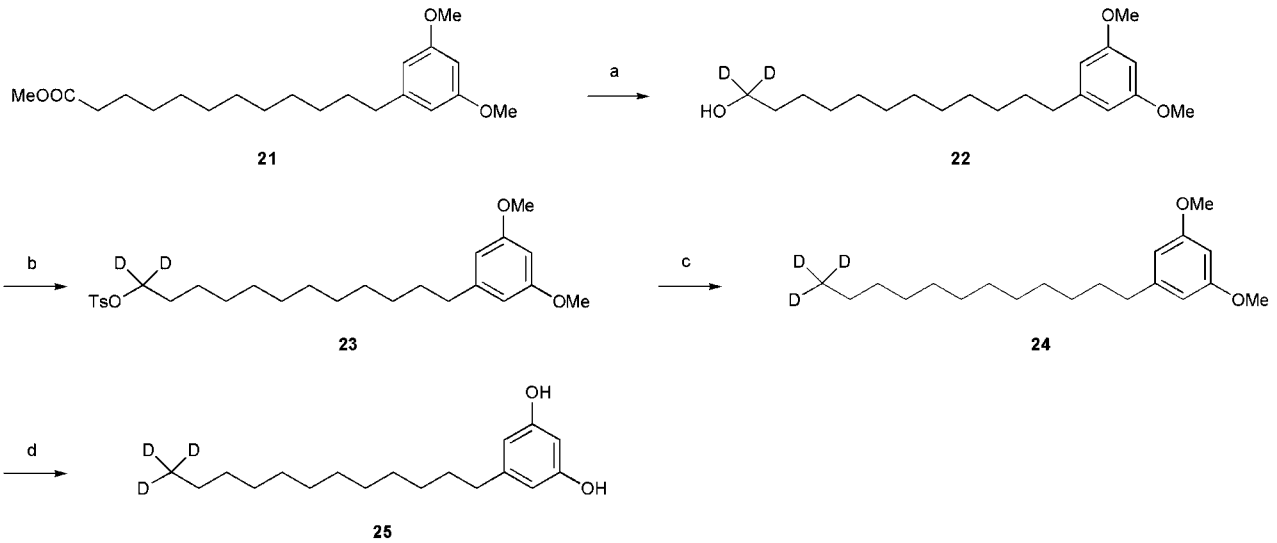
[1,1-$^2H_2$]-1-Bromooctadecane **5** (0.43 g, 1.3 mmol) and PPh_3 (0.4 g, 1.5 mmol) were refluxed in dry toluene (10 ml) under argon for 22 h at 130°C. The solution was allowed to cool down and evaporated. The product **6** was crystallized from Et_2O as a white powder (0.64 g, 84%), m.p. 96°C. ^1H NMR (200 MHz, $CDCl_3$) δ 0.80–0.85 (m, 3H, CH_3), 1.17–1.35 (m, 30H, CH_2), 1.50–1.60 (m, 2H, CH_2), 7.65–7.99 (m, 15H, ArH).

[1,2-$^2H_2$]-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. $^2H_1$-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 Et_2O (3 × 20 ml). Combined extracts were washed with brine and dried over Na_2SO_4. The product was purified by flash chromatography eluting with hexane/CH_2Cl_2 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-$^2H_4$]-1-(3,5-Dimethoxyphenyl)nonadecane 8

[1,2-$^2H_2$]-1-(3,5-Dimethoxyphenyl)-1-nonadecene **7** (0.025 g, 0.06 mmol) was stirred in CH_2Cl_2 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 ($^2H_2$, 10%, $^2H_3$, 30%, $^2H_4$, 60%, 0.022 g, 90%). ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.85–0.91 (m, 3H, CH_3), 1.13–1.38 (m, 30H, CH_2), 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-$^2H_4$]-1-(3,5-Dihydroxyphenyl)nonadecane 9

[1,1,2,2-$^2H_4$]-1-(3,5-Dimethoxyphenyl)nonadecane **8** (0.02 g, 0.05 mmol) was stirred with BBr_3 (0.44 ml, 1 M solution in CH_2Cl_2) in CH_2Cl_2 (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 $NaHCO_3$ and dried over Na_2SO_4. Flash chromatography on silica gel eluting with MeOH/EtOAc/CH_2Cl_2 1/2/7 gave **9** as a mixture of isotopologues ($^2H_2$, 10%, $^2H_3$, 40%, $^2H_4$, 50%, 0.015 g, 79%). ^{1}H NMR (200 MHz, D_6-acetone) δ 0.86–0.96 (m, 3H, CH_3), 1.20–1.35 (m, 30H, CH_2), 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 CH_2Cl_2 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 ($^2H_0$-$^2H_{15}$, $^2H_4$ and $^2H_5$ the main isotopologues) (0.51 g, 98%). A similar mixture ($^2H_0$-$^2H_{15}$) was obtained using Pd/$BaSO_4$ (0.1 g, 5% w/w) instead of Pd/C (0.51 g, 99%). ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.85–0.91 (m, 3H, CH_3), 1.15–1.40 (m, c.a. 24H, CH_2), 1.60–1.67 (m, 2H, CH_2), 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 Et_2O (3 × 30 ml). The combined extracts were dried over $MgSO_4$. Flash chromatography on silica gel eluting with hexane/acetone 1/1 gave **12** (0.41 g, 52%) as an amorphous solid. ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.83–0.90 (m, 3H, CH_3), 1.12–1.38 (m, 22H, CH_2), 1.40–1.55 (m, 2H, CH_2), 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); ^{13}C NMR (200 MHz, $CDCl_3$) 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 CH_2Cl_2 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 ($^2H_2$-$^2H_7$, $^2H_3$-$^2H_5$ as the main products, 0.13 g, 93%). ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.84–0.90 (m, 3H, CH_3), 1.12–1.37 (m, ca. 26H, CH_2), 1.50–1.56 (m, 2H, CH_2), 3.62 (t, 2H, $J = 6.6$ Hz, H-1).

Deuterated bromooctadecane 14

Concentrated HBr (15 ml), H_2SO_4 (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 $NaHCO_3$. Evaporation gave **14** as a pale yellowish powder, which was used in the next step without purification (isotopologues $^2H_2$-$^2H_7$, $^2H_3$-$^2H_5$ as the main products, 0.087 g, 77%). ^{1}H NMR (200 MHz, $CDCl_3$) δ 0.85–0.91 (m, 3H, CH_3), 1.13–1.38 (m, ca. 26H, CH_2), 1.79–1.89 (m, 2H, CH_2), 3.40 (t, 2H, $J = 6.6$ Hz, H-1).

Deuterated octadecyltriphenylphosphonium bromide 15

Deuterated bromooctadecane **14** (0.087 g, 0.26 mmol) and PPh_3 (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 Et_2O as a white powder (isotopologues $^2H_2$-$^2H_7$, $^2H_3$-$^2H_5$ as the main products, 0.11 g, 71%). ^1H NMR (200 MHz, $CDCl_3$) δ 0.82–0.87 (m, 3H, CH_3), 1.18–1.36 (m, ca. 26H, CH_2), 1.56–1.66 (m, 2H, CH_2), 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 Et_2O (3 × 20 ml). Combined extracts were washed with brine and dried over Na_2SO_4. The product was purified by flash chromatography eluting with hexane/CH_2Cl_2 2/3 to give a colourless oil (isotopologues $^2H_2$, 10%; $^2H_3$, 21%; $^2H_4$, 31%; $^2H_5$, 13%; $^2H_6$, 17%; $^2H_7$, 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 H_2 gas in CH_2Cl_2 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 $^2H_2$, 11%; $^2H_3$, 20%; $^2H_4$, 29%; $^2H_5$, 20%; $^2H_6$, 12%; $^2H_7$, 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 $MgSO_4$. 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 mmHg[18]). ^1H NMR (200 MHz, $CDCl_3$) δ 0.85–0.93 (m, 3H, CH_3), 1.31–1.55 (m, 6H, CH_2), 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), $PdCl_2$ (dppf) (0.016 g, 0.02 mmol) and 3,5-dimethoxyphenol trifluoromethanesulfphonate (0.19 g, 0.66 mmol) were added and the mixture refluxed for 2 h. Water was added and the product extracted with Et_2O (3 × 20 ml). The combined extracts were washed with brine and dried over Na_2SO_4. The product was purified by flash chromatography eluting with CH_2Cl_2/hexane 2/1 to give a colourless oil (0.024 g, 24%). ^1H NMR (200 MHz, $CDCl_3$) δ 0.88–0.94 (m, 3H, CH_3), 1.34–1.57 (m, 6H, CH_2), 1.75–1.85 (m, 2H, CH_2), 2.14–2.20 (m, 4H, CH_2), 2.66 (t, 2H, $J = 7.6$ Hz, CH_2Ar), 3.78 (s, 6H, OCH_3), 6.30 (t, 1H, $J = 2.2$ Hz, H-4), 6.36 (d, 2H, $J = 2.2$ Hz, H-2 and H-6); ^{13}C NMR (200 MHz, $CDCl_3$) 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^+.

Deuterogenation of 19

1-(3,5-Dimethoxyphenyl)-4-decyne (**19**) (0.024 g, 0.087 mmol) was stirred in CH_2Cl_2 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 $^2H_3$, 18%; $^2H_4$, 20%; $^2H_5$, 20%; $^2H_6$, 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-$^2H_2$]-12-(3,5-Dimethoxyphenyl)dodecan-1-ol 22

Methyl 12-(3,5-dimethoxyphenyl)dodecanoate **21** (0.075 g, 0.21 mmol) and $LiAlD_4$ (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 Na_2SO_4. Evaporation gave **22** as a white powder which was used in the next step without purification (0.070 g, 93%). ^1H NMR (300 MHz, $CDCl_3$) δ 1.26–1.35 (m, 16H, CH_2), 1.53–1.60 (m, 4H, CH_2), 2.54 (t, 2H, $J = 7.6$ Hz, CH_2Ar), 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′); ^{13}C NMR (300 MHz, $CDCl_3$) 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-$^2H_2$]-12-(3,5-Dimethoxyphenyl)dodecyl toluenesulphonate 23

[1,1-$^2H_2$]-12-(3,5-Dimethoxyphenyl)dodecan-1-ol **22** (0.06 g, 0.18 mmol), toluenesulphonyl chloride (0.041 g, 0.22 mmol) and Et_3N (0.04 ml) were stirred in CH_2Cl_2 (5 ml) at room temperature overnight. Water was added and the product extracted with CH_2Cl_2 (3 × 10 ml). Combined extracts were washed with brine and dried over Na_2SO_4. Flash chromatography on silica gel eluting with CH_2Cl_2 gave **23** (0.053 g, 62%). ^1H NMR (300 MHz, $CDCl_3$) δ 1.21–1.29 (m, 16H, CH_2), 1.53–1.64 (m, 4H, CH_2), 2.44 (s, 3H, CH_3), 2.54 (t, 2H, $J = 7.6$ Hz, CH_2Ar), 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); ^{13}C NMR (300 MHz, $CDCl_3$) 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-$^2H_3$]-1-(3,5-Dimethoxyphenyl)dodecane 24

[1,1-$^2H_2$]-12-(3,5-Dimethoxyphenyl)dodecyl toluenesulphonate **23** (0.050 g, 0.1 mmol) and $NaBD_4$ (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 Et_2O (3 × 10 m). The combined extracts were dried over Na_2SO_4. Flash chromatography on silica gel eluting with CH_2Cl_2 gave **24** (0.019 g, 63%). ^1H NMR (300 MHz, $CDCl_3$) δ 1.26–1.31 (m, 18H, CH_2), 1.53–1.64 (m, 2H, CH_2), 2.54 (t, 2H, $J = 7.8$ Hz, CH_2Ar), 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′); ^{13}C NMR (300 MHz, $CDCl_3$) 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-$^2H_3$]-1-(3,5-Dihydroxyphenyl)dodecane 25

[12,12,12-$^2H_3$]1-(3,5-Dimethoxyphenyl)dodecane **24** (0.016 g, 0.05 mmol) was stirred with BBr_3 (0.21 ml, 1 M solution in CH_2Cl_2) in CH_2Cl_2 (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 $NaHCO_3$ and dried over Na_2SO_4. Flash chromatography on silica gel eluting with MeOH/CH_2Cl_2 1/9 gave **25** (0.012 g, 80%). ^1H NMR (300 MHz, D_6-acetone) δ 1.20–1.35 (m, 18H, CH_2), 1.54–1.58 (m, 2H, CH_2), 2.44 (t, 2H, $J = 7.6$ Hz, CH_2Ar), 6.18 (s, 3H, H-2′, H-4′ and H-6′), 7.96 (s, 2H, OH); ^{13}C NMR (300 MHz, D_6-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 $^2H_2$-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.

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