First synthesis of [1,3,5-$^{13}C_3$]gallic acid

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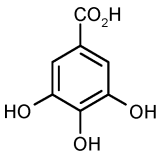
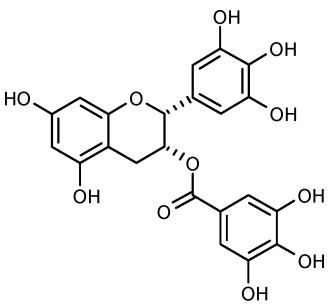
An efficient and high-yielding synthesis for [1,3,5-$^{13}C_3$]gallic acid from non-aromatic precursors is presented. [3,5-$^{13}C_2$]4*H*-Pyran-4-one was first prepared from the reaction between triethyl orthoformate and [1,3-$^{13}C_2$]acetone. The third ^{13}C-atom was introduced into the ring by reaction of the pyranone with diethyl [2-^{13}C]malonate. The resulting ethyl 4-hydroxy-[1,3,5-$^{13}C_3$]benzoate was brominated in the 3- and 5-positions to give ethyl 3,5-dibromo-4-hydroxy-[1,3,5-$^{13}C_3$]benzoate. Subsequent hydrolysis of the ester and substitution of the bromine atoms with hydroxyl groups was achieved under basic conditions in a single step to yield the desired [1,3,5-$^{13}C_3$]gallic acid. The synthesis of [2,6-$^{13}C_2$]4*H*-pyran-4-one is also presented to demonstrate the potential of the methodology for the regioselective placement of ^{13}C-atoms into benzene rings.

Introduction

Gallic acid **1a**, or 3,4,5-trihydroxybenzoic acid, is a phenolic plant metabolite which is biosynthesised *via* the shikimate pathway.[1] It is normally encountered in plant tissues in the ester form. Naturally occurring gallate esters include a group of catechins found in green tea, which have antioxidant properties.[2,3] The regular drinking of green tea has been associated with a reduced risk of several forms of cancer.[2,3] Epigallocatechin-3-gallate **2** is the most abundant catechin in green tea and has been shown to inhibit carcinogenesis.[2,3]



1a gallic acid



2 (-)-epigallocatechin-3-gallate

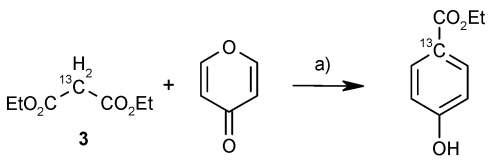
Ongoing biological studies into the absorption, metabolism and bioavailability of tea catechins require accurate quantification of the compounds in plasma samples. These samples are typically analysed by LC-MS or GC-MS assay methods. The accuracy and reproducibility of these techniques are greatly improved by the use of stable isotope labelled standards. Our aim was to prepare stable isotope labelled versions of gallic acid for incorporation into tea catechins such as **2**.

Previous syntheses of isotopically labelled gallic acid include the preparation of [2,6-$^2H_2$]gallic acid by Tuck *et al. via* deuterium exchange.[4] However, in the context of GC-MS and LC-MS analyses, deuterated standards are often not suitable, as the deuterium atoms can exchange out of the compound during sample preparation and analysis. Also, deuterium exchange methods tend to produce mixtures of deuterated species which give broad MS envelopes. HPLC retention times can also be affected by the presence of deuterium.[5,6]

Two routes to [*carboxy*-^{14}C]gallic acid have been reported, by Zeng *et al.*[7] and Schildknecht *et al.*,[8] employing $^{14}CO_2$ as the labelled species, but no syntheses of any ring-labelled versions have been reported.

In order to avoid the inherent disadvantages of deuterium labelled internal standards, we chose to prepare ring-labelled versions of gallic acid containing multiple ^{13}C-atoms. Our strategy was to construct the aromatic ring using a pyranone intermediate which could be prepared from simple acyclic precursors. ^{13}C-Labelled acyclic precursors are more widely available and less expensive than uniformly labelled aromatic compounds. This approach would allow regiospecific introduction of ^{13}C-atoms into the aromatic ring and thus allow the preparation of a number of isotopically labelled versions (isotopomers) of gallic acid.

It was decided to adopt a synthetic strategy utilising some chemistry originally developed by Steglich and co-workers.[10,11] They employed the reaction of diethyl [2-^{13}C]malonate **3** with 4*H*-pyran-4-one **4**, under basic conditions to synthesise ethyl 4-hydroxy-[1-^{13}C]benzoate **5** (Scheme 1). However, in order to exploit this chemistry for our purposes it was necessary to incorporate further ^{13}C-atoms into the 4*H*-pyran-4-one and so the synthesis of this material from ^{13}C-labelled precursors was developed.



Scheme 1 *Reagents and conditions*: (a) ^{t}BuOH, KO^{t}Bu (1.1 eq.), then HCl, H_2O, 80%.

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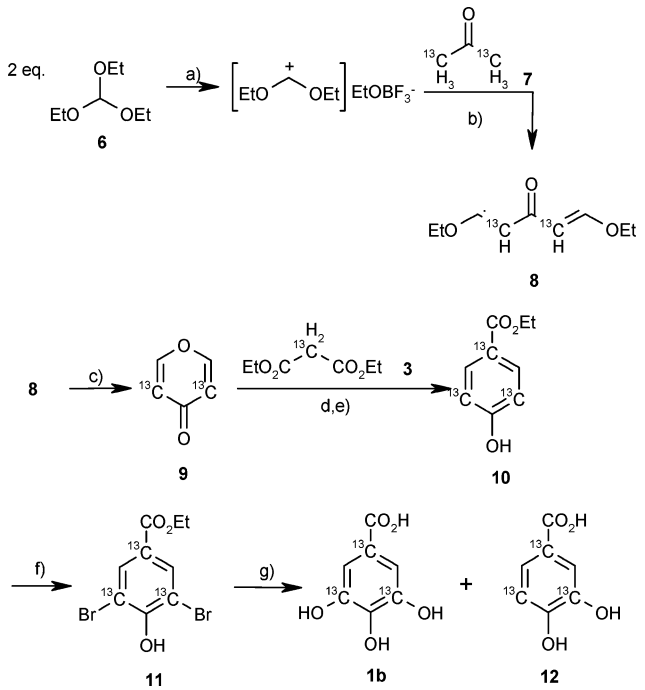
Results and discussion

Our synthesis of [1,3,5-$^{13}C_3$]gallic acid **1b** (Scheme 2) commenced with triethyl orthoformate **6** and [1,3-$^{13}C_2$]acetone **7**, using a similar procedure to that developed by Hobuss and co-workers.[9] Triethyl orthoformate **6** was first treated with boron trifluoride diethyl etherate, followed by the addition of the ^{13}C-labelled acetone **7** and *N,N*-diisopropylethylamine. Optimisation studies were carried out, varying the number of equivalents of triethyl orthoformate and acetone used in the reaction. It was found that the stoichiometries used by Hobuss and co-workers[9] (1 equivalent acetone, 6 equivalents triethyl orthoformate) were not the optimum conditions for the reaction, and that our optimised conditions (1 equivalent acetone, 2 equivalents triethyl orthoformate) gave a much cleaner reaction, allowing the intermediate enone **8** to be used *in situ* without purification. Following cyclisation under acidic conditions, the [3,5-$^{13}C_2$]4*H*-pyran-4-one **9** was isolated from the aqueous phase, and not the organic phase as implied in the original publication,[9] in excellent yield. It was observed during isolation of the pyranone that it is sensitive to light, heat and acid, so our procedure accounts for these factors.



Scheme 2 *Reagents and conditions:* (a) $BF_3{\cdot}OEt_2$ (6 eq.), DCM; (b) $^{i}Pr_2NEt$ (6 eq.) quant.; (c) EtOH, HCl, H_2O, quant.; (d) $^{t}BuOH$, $KO^{t}Bu$ (1.1 eq.); (e) HCl, H_2O, 74%; (f) AcOH, NaOAc, Br_2 (4.2 eq.) 99%; (g) NaOH, H_2O, $CuSO_4{\cdot}5H_2O$ (0.6 eq.), 48% **1b** (6% 3,4-dihydroxy-[1,3,5-$^{13}C_3$]benzoic acid **12**).

The synthesis of [2,6-$^{13}C_2$]4*H*-pyran-4-one was also carried out using our optimised procedure. This was achieved from the reaction of unlabelled acetone and triethyl [^{13}C]orthoformate, to yield the labelled pyranone in excellent yield. This demonstrates the potential of our procedure for the regioselective placement of ^{13}C-atoms within the pyranone ring. In total, it is possible to synthesise seven different isotopomers of 4*H*-pyran-4-one, with from one to five ^{13}C-atoms, by using varying combinations of the commercially available ^{13}C-labelled versions of the acetone and/or triethyl orthoformate starting materials.

The development and optimisation of Steglich's method,[10,11] using our ^{13}C-labelled 4*H*-pyran-4-one, led to the isolation of ethyl 4-hydroxy-[1,3,5-$^{13}C_3$]benzoate **10** in good yield, with no requirement for purification. This involved the reaction of the pyranone **9** with diethyl [2-^{13}C]malonate **3** in the presence of potassium *tert*-butoxide as base. Aromatisation and loss of carbon dioxide occurred during the acidic work-up to give *para*-substituted phenol **10** with ^{13}C-labels in three pre-determined positions in the ring.

Bromination of **10** to give ethyl 3,5-dibromo-4-hydroxy-[1,3,5-$^{13}C_3$]benzoate **11** was first attempted using dichloromethane and an excess of bromine at 0 °C.[10,11] However, this was unsuccessful, giving only traces of the mono-bromo compound and none of the desired di-bromo compound. It was found that the use of an excess of bromine in acetic acid with sodium acetate at room temperature was an extremely fast, efficient, and high yielding reaction, with only the desired di-bromo compound **11** being formed. Once again, purification was not required.

It had been shown that the mono-bromo compound could firstly be hydrolysed to the acid using sodium hydroxide, followed by substitution of the bromine atoms with hydroxyl groups using activated copper and copper sulfate in aqueous potassium hydroxide.[12,13] However, when this was attempted using the di-bromo compound, only 4-hydroxybenzoic acid was recovered from the reaction. Various conditions were investigated and it was found that the two steps could be combined into one single step using degassed aqueous sodium hydroxide and aqueous copper sulfate to give both hydrolysis of the ester and substitution of the bromine atoms to hydroxyl groups in a single step. This gave the desired [1,3,5-$^{13}C_3$]gallic acid **1b** in good yield with a small amount of 3,4-dihydroxy-[1,3,5-$^{13}C_3$]benzoic acid **12** as by-product. Purification was only carried out at this stage, and gave the final product **1b** in an overall yield of 35% after column chromatography.

Experimental

General comments

NMR spectra were recorded on a Varian Gemini 2000 (^{1}H 300 MHz, ^{13}C 75.45 MHz) or a Bruker Avance 400 (^{1}H 400 MHz, ^{13}C 100 MHz) spectrometer. ^{13}C NMR spectra were recorded using the PENDANT or DEPTQ sequence and internal deuterium lock. Chemical shifts (δ) in ppm are given relative to Me_4Si, coupling constants (J) are given in Hz. IR spectra were recorded on a Perkin-Elmer series 1420 FT IR spectrophotometer. The samples were prepared as Nujol mulls or thin films between sodium chloride discs and recorded in cm^{-1}. Low resolution and high resolution EI and CI mass spectra were recorded on a Micromass LC-T (time-of-flight). Melting points were recorded on an Electrothermal melting point apparatus and are uncorrected. Analytical TLC was carried out on Merck 5785 Kieselgel $60F_{254}$ fluorescent plates. The components were observed under ultraviolet light (254 nm). Flash chromatography was performed according to the procedure of Still *et al.*[14] using silica gel of 35–70 μ particle size. *tert*-Butanol was dried by stirring with 3 Å molecular sieves overnight, followed by filtration. All chemicals were used as received unless otherwise indicated.

[3,5-$^{13}C_2$]4*H*-Pyran-4-one (9)[9]. To triethyl orthoformate (4.74 g, 5.33 mL, 32 mmol) at −30 °C (acetonitrile and dry ice) under argon, was added drop-wise over 15 min, a solution of $BF_3{\cdot}OEt_2$ (12.14 mL, 96 mmol) in DCM (13 mL). The reaction mixture was warmed to 0 °C with stirring, and stirred for 20 min to give a slurry of diethoxycarbenium fluoroborate. The mixture was cooled to −78 °C and [1,3-$^{13}C_2$]acetone (1.0 g, 14 mmol) added. $^{i}Pr_2NEt$ (16.7 mL, 96 mmol) was then added drop-wise over 30 min with efficient stirring and the resulting reaction mixture stirred at −78 °C for 2.5 h. After that time, the reaction mixture was poured into conc. $NaHCO_3$ solution (120 mL), and stirred vigorously for 10 min at room temperature. The resulting mixture was extracted with DCM (3 × 50 mL). The combined organic layers were washed successively with ice cold 1 M H_2SO_4 (17.5 mL), and cold water (2 × 50 mL) and dried ($MgSO_4$). Removal of the solvent at reduced pressure gave the crude enone product **8** as a brown oil (2.40 g, quant.). δ_H (300 MHz, $CDCl_3$) 7.52 (2H, dd, *J* 12.4, 6.7 Hz, -CH-), 5.59 (2H, dd, *J* 159.3, 12.4 Hz, -^{13}CH-), 3.88 (4H, q, *J* 7.1 Hz, -CH_2-), and 1.28 (6H, t, *J* 7.1 Hz, -CH_3); δ_C (75.45 MHz, $CDCl_3$) 163.3 (t, *J* 41 Hz, -CO-), 105.6 (enhanced, -CH-), 62.5 (–CH_2-), and 14.4 (–CH_3).

A solution of the crude enone **8** (2.75 g, 16 mmol) in ethanol (80 mL) and 10% aqueous HCl (8 mL) was heated at 80 °C for 24 h. After this time, the solvents were removed at reduced pressure and the resulting residue dissolved in water (50 mL). The aqueous layer was extracted with toluene (3 × 30 mL), and the combined organic layers washed with water (2 × 30 mL). After filtering the aqueous phase to remove any solids, it was treated with decolourising charcoal and filtered through celite. Removal of the solvent from the aqueous phase at reduced pressure (at 40 °C) gave [3,5-$^{13}C_2$]4*H*-pyran-4-one **9** as an off-white solid (1.37 g, quant.). Mp 32–32.5 °C (lit unlabelled[9] 32.5 °C); *m*/*z* (CI) 99 [$(MH)^+$, 100%]; HRMS (CI) [Found: $(MH)^+$, 99.0357, $^{12}C_3{}^{13}C_2H_5O_2$ requires 99.0357]; δ_H (300 MHz, DMSO-d_6) 8.13 (2H, ddd, *J* 6.5, 4.8, 2.1 Hz, C(2,6)H), and 6.29 (2H, dd, *J* 171.7, 6.5 Hz, ^{13}C(3,5)H); δ_C (75.45 MHz, DMSO-d_6) 177.1 (t, *J* 54 Hz, -CO-), 156.9 (d, *J* 72 Hz, C-2, 6), and 117.3 (enhanced, C-3,5); ν_{max}/cm^{-1} 1619 (C=O), 1590 (C=C), 1326 (CH), 1091 (C-O), 918 and 848 (CH).

[2,6-$^{13}C_2$]4*H*-Pyran-4-one (14)[9]. To triethyl [^{13}C]orthoformate (1 g, 1.12 mL, 6.7 mmol) at −30 °C (acetonitrile and dry ice) under argon, was added drop-wise over 15 min, a solution of $BF_3{\cdot}OEt_2$ (2.54 mL, 20.1 mmol) in DCM (3 mL). The reaction mixture was warmed to 0 °C with stirring, and stirred for 20 min to give a slurry of diethoxycarbenium fluoroborate. The mixture was then cooled to −78 °C and acetone (0.24 mL, 3.35 mmol) was added. $^{i}Pr_2NEt$ (3.5 mL, 20.1 mmol) was then added drop-wise over 30 min with efficient stirring and the resulting reaction mixture stirred at −78 °C for 2.5 h. After that time, the reaction mixture was poured into conc. $NaHCO_3$ solution (30 mL), and stirred vigorously for 10 min at room temperature. The resulting mixture was extracted with DCM (3 × 15 mL). The combined organic layers were washed successively with ice cold 1 M H_2SO_4 (4.2 mL), and cold water (2 × 15 mL) and dried ($MgSO_4$). Removal of the solvent at reduced pressure gave the crude enone **13** as a brown oil (0.58 g, quant.). δ_H (300 MHz, $CDCl_3$) 7.52 (2H, dd, *J* 181.4, 12.5 Hz, -^{13}CH-), 5.57 (2H, dd, *J* 12.5, 3.2 Hz, -CH-), 3.88 (4H, dq, *J* 7.0, 2.9 Hz, -CH_2-), and 1.28 (6H, t, *J* 7.0 Hz, -CH_3); δ_C (75.45 MHz, $CDCl_3$) 162.8 (-CO-), 161.4 (enhanced, -CH-), 105.6 (d, *J* 79 Hz, -CH-), 67.2 (-CH_2-), and 14.6 (-CH_3).

A solution of the crude enone (0.58 g, 3.35 mmol) in ethanol (20 mL) and 10% aqueous HCl (2 mL) was heated at 80 °C for 24 h. After this time, the solvents were removed at reduced pressure and the resulting residue dissolved in water (15 mL). The aqueous layer was extracted with toluene (3 × 15 mL), and the combined organic layers washed with water (2 × 15 mL). After filtering the aqueous phase to remove any solids, it was treated with decolourising charcoal and filtered through celite. Removal of the solvent from the aqueous phase at reduced pressure (at 40 °C) gave [2,6-$^{13}C_2$]4*H*-pyran-4-one **14** as an off-white solid (0.304 g, 92%). Mp 33–33.5 °C (lit unlabelled[9] 32.5 °C); *m*/*z* (CI) 99 [$(MH)^+$, 100%]; HRMS (CI) [Found: $(MH)^+$, 99.0360, $^{12}C_3{}^{13}C_2H_5O_2$ requires 99.0357]; δ_H (300 MHz, CD_3OD) 8.20 (2H, dm, *J* 202 Hz, ^{13}C(2,6)H), and 6.33 (2H, m, C(3,5)H); δ_C (75.45 MHz, CD_3OD) 156.8 (enhanced, C-2, 6), and 117.9 (s, C-3,5); ν_{max}/cm^{-1} 1623 (C=O), 1577 (C=C), 1325, 923 and 827 (CH).

Ethyl 4-hydroxy-[1,3,5-$^{13}C_3$]benzoate (10). A solution of [3,5-$^{13}C_2$]4*H*-pyran-4-one **9** (1.57 g, 16 mmol) and diethyl [2-^{13}C]malonate **3** (2.34 g, 15 mmol) in dry ^{t}BuOH (25 mL) was heated to reflux under argon. KOBu^{t} (16 mmol, 16 mL of a 1 M solution in ^{t}BuOH) in dry ^{t}BuOH (30 mL) was added drop-wise. The resulting mixture was heated under reflux for 3 h, after which time HCl (1 M, 30 mL) was added. The reaction solution was heated under reflux for a further 1 h, the solvent was then removed at reduced pressure, and water (40 mL) added. The aqueous phase was extracted with diethyl ether (3 × 30 mL). The combined organic phase was washed with water (2 × 30 mL), brine (30 mL), dried ($MgSO_4$) and solvent removed at reduced pressure to give **10** as an orange solid (1.88 g, 74%). Mp 113–113.5 °C (lit unlabelled[8] 112–114 °C); *m*/*z* (CI) 170 [$(MH)^+$, 100%], 142 [$(M - Et)^+$, 18]; HRMS (CI) [Found: $(MH)^+$, 170.0808, $^{12}C_6{}^{13}C_3H_{10}O_3$ requires 170.0809]; δ_H (400 MHz, $CDCl_3$) 7.97 (2H, br d, *J* 8.7 Hz, C(2,6)H), 6.88 (2H, dm, *J* 159.2 Hz, ^{13}C(3,5)H), 4.36 (2H, q, *J* 7.3 Hz, -CH_2-), and 1.39 (3H, t, *J* 7.3 Hz, -CH_3); δ_C (100 MHz, $CDCl_3$) 169.7 (-CO-), 161.4 (C-4), 131.8 (dt, *J* 59, 6 Hz, C-2,6), 123.0 (enhanced, t, *J* 2 Hz, C-1), 115.1 (enhanced, d, *J* 2 Hz, C-3,5), 60.9 (-CH_2-), and 14.4 (-CH_3); ν_{max}/cm^{-1} 3207 (OH), 1714 (C=O), 1667 and 1569 (ArH), 1277 (OH), 1229, 1165, 1098 and 1016 (C–O), 840 and 762 (CH).

Ethyl 3,5-dibromo-4-hydroxy-[1,3,5-$^{13}C_3$]benzoate (11). To a solution of ethyl [1,3,5-$^{13}C_3$]4-hydroxybenzoate **10** (0.878 g, 5.2 mmol) and sodium acetate (1.94 g, 24 mmol) in AcOH (20 mL), was added drop-wise over 20 min a solution of bromine (3.54 g, 22 mmol) in AcOH (5 mL) at room temperature. The reaction mixture was stirred for 1.5 h at room temperature. After the addition of water (40 mL) to the reaction mixture, the aqueous phase was extracted with DCM (3 × 40 mL). The combined organic layers were washed with sodium metabisulfite solution (2 × 30 mL), water (30 mL), brine (30 mL) and dried ($MgSO_4$). Removal of the solvent at reduced pressure gave the di-bromo product **11** as an orange/yellow solid (1.68 g, 99%). Mp 150–150.5 °C (lit unlabelled[15] 108–108.5 °C); *m*/*z* (CI) 328 [$(MH^{79}Br^{81}Br)^+$, 100%], 326 [$(MH^{79}Br_2)^+$, 30], 330 [$(MH^{81}Br_2)^+$, 27]; HRMS (CI) [Found: $(MH)^+$, 327.8998, $^{12}C_6{}^{13}C_3H_8O_3{}^{79}Br^{81}Br$ requires 327.8999]; δ_H (400 MHz, $CDCl_3$) 8.16 (2H, dt, *J* 2.8, 1.7 Hz, C(2,6)H), 4.37 (2H, q, *J* 7.1 Hz, -CH_2-), and 1.40 (3H, t, *J* 7.1 Hz, -CH_3); δ_C

(100 MHz, $CDCl_3$) 164.7 (-CO-), 153.1 (C-4), 133.6 (ddd, *J* 69, 59, 4 Hz, C-2,6), 125.1 (enhanced, C-1), 109.6 (enhanced, C-3,5), 61.5 (-CH_2-), and 14.3 (-CH_3); ν_{max}/cm^{-1} 3313 and 2987 (OH), 1692 (C=O), 1458 (ArH), 1367 and 1281 (OH), 1240, 1212, 1112 and 1016 (C-O), 903 and 758 (CH), and 719 (C-Br).

[1,3,5-$^{13}C_3$]Gallic acid or 3,4,5-trihydroxy-[1,3,5-$^{13}C_3$]benzoic acid (1b). A solution of NaOH (7.0 g, 0.17 mol) in water (50 mL) was stirred under reduced pressure for 90 min. A solution of $CuSO_4$ (0.44 g, 2.75 mmol) was then added to the basic solution and stirring continued under reduced pressure for an additional 4 h. The aqueous solution was transferred by cannula to a flask containing ethyl [1,3,5-$^{13}C_3$]3,5-dibromo-4-hydroxybenzoate **11** (1.5 g, 4.6 mmol). After allowing the blue suspension to stir at room temperature under argon for 90 min, the mixture was heated at reflux at 110 °C for 18 h, cooled, and acidified with HCl. The aqueous mixture was extracted with both diethyl ether (3 × 40 mL) and ethyl acetate (3 × 40 mL). The combined organic extracts were washed with brine (2 × 40 mL) and dried ($MgSO_4$). Removal of the solvent at reduced pressure gave crude product (0.6 g, 75%) which was a mixture of [1,3,5-$^{13}C_3$]gallic acid and 3,4-dihydroxy-[1,3,5-$^{13}C_3$]benzoic acid (10 : 1). Purification by column chromatography (silica gel, petroleum ether–ethanol, 10 : 1), Rf = 0.356, gave the pure product **1b** as an off-white solid (0.38 g, 48%). Mp 262–263 °C (lit unlabelled[16] 258–260 °C); *m/z* (CI) 174, [$(MH)^+$, 100%], 156 [$(M - OH)^+$, 6]; HRMS (CI) [Found: $(MH)^+$, 174.0397, $^{12}C_4{}^{13}C_3H_7O_5$ requires 174.0394]; δ_H (300 MHz, acetone-d_6) 8.10 (2H, s, OH), 7.90 (1H, s, OH), and 7.02 (2H, dd, *J* 3.8, 1.9 Hz, C(2,6)H); δ_C (75.45 MHz, acetone-d_6) 151.6 (-CO-), 146.0 (enhanced, C-3,5), 140.5 (C-4), 122.0 (enhanced, C-1), 110.2 (C-2,6); ν_{max}/cm^{-1} 3272 and 2988 (OH), 1643 (C=O) and 1574 (ArH), 1410, 1310 and 1262 (OH), 1219 and 1023 (C–O), 865, 760 and 731 (CH). Data for 3,4,-dihydroxy-[1,3,5-$^{13}C_3$]benzoic acid **12**: (41 mg, 6%). Mp 183.5–184 °C (lit unlabelled[17] 189 °C); *m/z* (CI) 158 [$(MH)^+$, 100%], 157 [M^+, 7]; HRMS (CI) [Found: $(MH)^+$, 158.0445, $^{12}C_4{}^{13}C_3H_7O_4$ requires 158.0445]; δ_H (300 MHz, acetone-d_6) 7.40 (1H, m, C(2)H), 7.34 (1H, dm, *J* 8.5 Hz, C(6)H), and 6.77 (1H, ddt, *J* 159.6, 8.5, 6.9 Hz, C(5)H); δ_C (75.45 MHz, acetone-d_6) 145.5 (enhanced, C-3), 123.1 (enhanced, C-1), 115.7 (enhanced, C-5); ν_{max}/cm^{-1} 3280 and 2990 (OH), 1656 (C=O), 1407 and 1275 (OH), 1231, 1113, and 1025 (C-O), 895 and 755 (CH).

Conclusions

It can therefore be concluded that a fast, efficient and high yielding route to the synthesis of [1,3,5-$^{13}C_3$]gallic acid has been developed starting from commercially available ^{13}C-labelled acyclic building blocks. Our route allows the regioselective placement of ^{13}C-atoms within the aromatic ring, demonstrated by the synthesis of both [2,6-$^{13}C_2$]4*H*-pyran-4-one and [3,5-$^{13}C_2$]4*H*-pyran-4-one; two of the possible seven ^{13}C-labelled isotopomers of the pyranone which could be synthesised using our approach. Also, the need for the minimum amount of purification, and therefore the minimum amount of losses of ^{13}C-labelled material, has been met, with purification only being required in the final step.

Acknowledgements

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