Synthesis of ring-^{14}C-labelled 4(3′, 6′-dimethyl-3′-heptyl)-phenol

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Summary

Ring-^{14}C-labelled 4(3′-, 6′-, dimethyl-3′-,heptyl)-phenol (NP), an isomer of paranonyl phenol was synthesized for use in aquatic toxicity and metabolism studies. A very efficient method involving alkylation of ring-^{14}C-labelled anisole and tertiary alkyl bromide (3-bromo-3,6-dimethyl heptane) with AlCl$_3$ followed by cleavage of the resultant ether with BBr$_3$ was used giving a 24% yield ring labelled ^{14}C-NP (specific activity: 16.205 MBq/mg and purity > 95% by HPLC) after TLC separation. Copyright © 2001 John Wiley & Sons, Ltd.

Key Words: Nonyl phenol; branched isomer; synthesis; [14]C

Introduction

Isomeric nonyl phenols (NPs) are a group of compounds used in industry to make alkylphenol polyethoxylates (APEs) for the manufacture of non-ionic surfactants, antioxidants, detergents, polymers, etc.[1–3] The APEs eventually find their way into drainage canals and sewage plants as waste where they degrade and release more NPs and

their derivatives. The NPs and their derivatives are persistent in the aquatic environment and are known to be toxic and have negative estrogenic effects on aquatic organisms.[4–6] An industrially produced NP is composed of a mixture of predominantly branched- and straight-chain nonyl derivatives of phenol containing more than 18 isomers.[1,2] The isomerization arises due to uncontrollable chemical reactions during manufacture resulting in a product mixture composed of 90% para-nonyl phenol (both branched and straight chain) isomers, 5–9% dinonyl phenols and small amounts of the ortho-isomers. These isomers have been identified in various NP products by GC–MS and NMR but have not been obtained as single compounds for metabolism studies and it is therefore not known which of the isomers are responsible for the toxic and estrogenic effects.[1,2,7] A few isomers such as the mixture of radiola-belled 4(2′-nonyl)-phenol, 4(3′-nonyl)-phenol and 4(4′-nonyl)-phenol have been synthesized by alkylation of ^{14}C-RUL-phenol with 1-nonene.[1] Meldhal et al.[1] expressed difficulty in getting a single 4-n-nonyl phenol due to the relative instability of the primary nonyl carbocation and the presence of different branched isomers in the alkene starting material. Ortho-alkylation also occurred in minor amounts in their synthesis.

In our synthesis, we obtained pure ring-^{14}C-labelled 4(3′-6′-dimethyl-3′-heptyl)-phenol (**4**) containing only traces of the ortho-product, 2(3′-, 6′-dimethyl-3′-heptyl)-phenol.

Results and discussion

The synthesis involved Friedel–Crafts alkylation of (ring-^{14}C)-anisole (**1**) and 3-bromo-3,6-dimethyl heptane (**2**) (synthesized earlier in our laboratory) with $AlCl_3$ followed by cleavage of the methoxy bond of the resultant ether (**3**) with BBr_3 as shown by the following reaction scheme (Scheme 1).[8–10]

The first reaction step was achieved easily and a high yield (97.6%) of the crude product (3) was obtained. This was left in the n-hexane solution since the second step could be achieved in the same solvent. The second reaction step was rather difficult to achieve and an optimum reaction time of 2.5 h had to be observed to avoid cleavage of the phenol-alkyl bond of the NP produced.

(i)= AlCl$_3$, < 20 $^\circ$C
(ii)= BBr$_3$, hexane, 0 $^\circ$C
*= radiolabel position
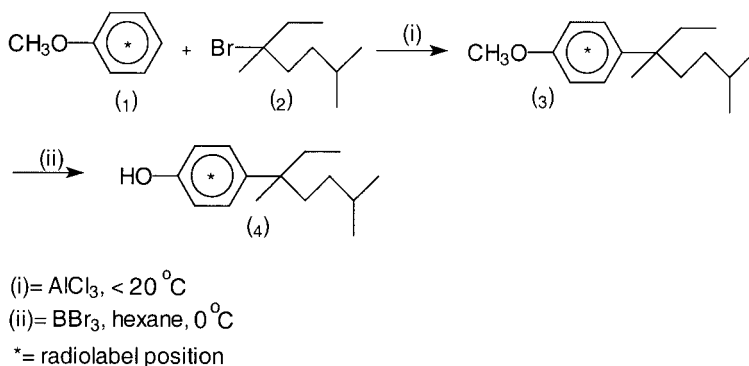
Scheme 1.

Experimental

TLC was done on Silica gel 60 F$_{254}$ (5 × 20 cm) and separation on Silica gel 60 F$_{254}$, 2 mm (20 × 20 cm); solvent: *n*-hexane:acetone (3:1 (vol)), viewed under UV light of 254 nm wavelength. Radioactive spots were also located by Berthold Automatic TLC-Linear Scanner Trace Master 20. Radioactivity was quantified by Packard Tri-Carb Liquid Scintillation Analyzer model 2100 TR. HPLC was performed on Merck Hitachi system (L-6200 A pump, L-4000 UV detector and D-2500 Chromato-Integrator) fitted to a Ramona-D radioactive detector, and column: RP 18, 10 cm long, 5 mm i.d.; solvent: 60% methanol in water; isocratic conditions. The GC–MS determination was done on GC HP 5890 Series II and MS Finnigan SSQ 7000; column: J&W DB-5 ms, 60 m, ID 0.25 mm, film thickness 0.10 μm. The temperature program: 60°C (1.5 min); 7.5°C/min to 260°C (12 min), injection temp: 250°C, 1 μl splitless injection; Carrier gas: helium pressure 25 psi. MS conditions: full scan from mass 50 to 500, cycle time 0.7 s, ionization mode: EI, 70 eV, ion source temperature: 150°C. The (ring-^{14}C)-anisole ($\underline{1}$) obtained from International Isotopes, Munich, was 98% pure (by HPLC) and had a specific activity of 7.955 MBq/mg.

4 (3′-, 6′-, dimethyl-3′-heptyl)-methoxybenzene ($\underline{3}$)

Twenty-five millilitres *n*-hexane solution of the ^{14}C-anisole (89.9 mg, 1.112 mCi/ml) was placed in a 50 ml two-necked reaction flask, with a magnetic stirrer and connected to a reflux condenser. Then AlCl$_3$

(6.36 mg) was added while stirring. The tertiary alkyl bromide (3-bromo-3,6-dimethyl heptane (2); 106 μl, density: 1.0909 g/cm^3) was added and the reaction mixture maintained below 20°C in an ice/water bath overnight. The next day the reaction was chilled by adding a few grams of ground ice and then 20 ml distilled, shaken and the organic phase separated in a separatory funnel. The phases were analysed by HPLC and aliquots taken for liquid scintillation counting. Very little radioactivity was detected in the water phase as most of the substance was in the *n*-hexane phase. The organic phase was washed with 20 ml 10% NaOH solution, washed again with 20 ml distilled water, until neutral, and dried with anhydrous sodium sulfate. It was then filtered and analyzed by LSC. Total radioactivity of 26.1 mCi was found in this product, representing 97.6% recovered radioactivity. This sample was also analyzed by TLC, HPLC and GC-MS and compared to a standard sample which had been earlier prepared and characterized by NMR and IR. It was found to be mainly the ether product, 4(3′-, 6′-, dimethyl-3′-heptyl)-methoxybenzene (3). From this crude product (3), 8 ml (containing 6.5 mCi) was used for the second step of the reaction.

4 (3′-, 6′-dimethyl-3′-heptyl)-phenol (4)

Eight millilitres of (3) (containing 6.5 mCi of crude substance) was placed in a 50 ml two-necked reaction flask, with stirrer and fitted with a reflux condenser. The volume was adjusted to 10 ml by adding more *n*-hexane. While cooling in an ice bath and stirring, 6 drops of BBr$_3$ were pipetted in and the reaction left to proceed for 1.5 h. The 10 ml distilled water was added and the mixture was left to react for a further 1 h. The aqueous and organic phases were then separated in a separatory funnel, the aqueous phase washed twice with 20 ml *n*-hexane and the total radioactivity determined in the two phases. It was found that 2.8 mCi was still left in the water phase and 2.5 mCi in the *n*-hexane phase since the product was more soluble in water than in *n*-hexane. The product was therefore extracted from the water phase with CH$_2$Cl$_2$, by first making the aqueous solution alkaline with a small amount of 10% NaOH (testing with pH indicator paper), extracting with *n*-hexane and then making it acidic with a few drops of dilute HCl solution and then extracting with CH$_2$Cl$_2$. The total combined crude product in *n*-hexane and in CH$_2$Cl$_2$ extracts was found to contain 4.7 mCi (72.3% of starting material) of the substance (4). The compound was confirmed by TLC

(R_f: 0.43), HPLC (R_t: 4.5) and GC–MS, using a normal standard prepared earlier in the laboratory. The solvent was removed at low temperature under low vacuum rotary evaporation and the remainder redissolved in a small volume of CH_2Cl_2. This crude product was spotted on preparative TLC plates, developed in *n*-hexane : acetone solvent and the spots located by UV light and TLC scanner. The separated compound was scrapped from the plate and dissolved in CH_2Cl_2. The solvent was then removed by rotary evaporation to yield a small quantity of brown liquid which was redissolved in 5 ml methanol for future use. After separation, a 24% yield of the compound (4) (containing 1.56 mCi, with purity > 95%) was obtained.

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