Synthesis of 4-Quinolone *N*-Oxides via Controlled Partial Hydrogenation of 2-Nitrobenzoyl Enamines

Yordanka Mollova-Sapundzhieva [1], Francisco Alonso [2], Plamen Angelov [1,*] and Paraskev Nedialkov [3]

[1] Department of Organic Chemistry, University of Plovdiv Paisii Hilendarski, 24 Tsar Asen Str., 4000 Plovdiv, Bulgaria

[2] Instituto de Síntesis Orgánica and Departamento de Química Orgánica, Facultad de Ciencias, Universidad de Alicante, Apdo. 99, Alicante, 03080, Spain.

[3] Department of Pharmacognosy, Faculty of Pharmacy, Medical University of Sofia, 2 Dunav Str., 1000 Sofia, Bulgaria

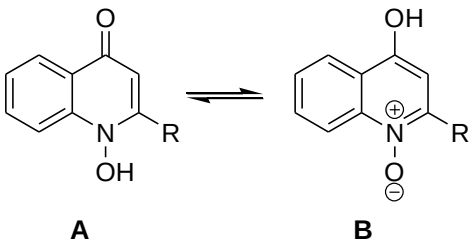
* Correspondence: angelov@uni-plovdiv.bg; Tel.: +359-32-261349

Abstract: An operationally simple procedure for the synthesis of 2-alkyl-4-quinolone *N*-oxides, relying on controlled platinum-catalyzed partial hydrogenation of 2-nitrobenzoyl enamines, has been demonstrated. The *P. aeruginosa* metabolite 2-heptyl-4-quinolone-*N*-oxide (HQNO) and four analogous products have been prepared in good yield and high purity by this method.

Keywords: *N*-hydroxy-4-quinolones, 4-hydroxyquinoline *N*-oxides, HQNO, β-enaminones, heterogeneous catalytic hydrogenation, platinum.

Introduction

The 2-Alkyl-4(1*H*)-quinolones (AQs) are a class of quorum-sensing signal molecules, which coordinate the production of virulence factors in *P. aeruginosa* and related species [1]. The function of their *N*-oxides (AQNOs) on the other hand is that of toxins and weapons in the interspecies competition [2]. Both, AQs and AQNOs, can exist in two tautomeric forms (Scheme 1) and for this reason they are also referred to as 4-hydroxyquinolines and 4-hydroxyquinoline *N*-oxides, respectively. Although the oxidized 4-quinolone tautomer is actually a *N*-hydroxy derivative (Scheme 1, A), the term "4-quinolone *N*-oxide" has gained widespread acceptance as a universal reference to this type of compounds, disregarding the precise tautomeric form.



Scheme 1. *Tautomeric forms of AQNOs: N-hydroxy-4-quinolone (**A**) and 4-hydroxyquinoline N-oxide (**B**).*

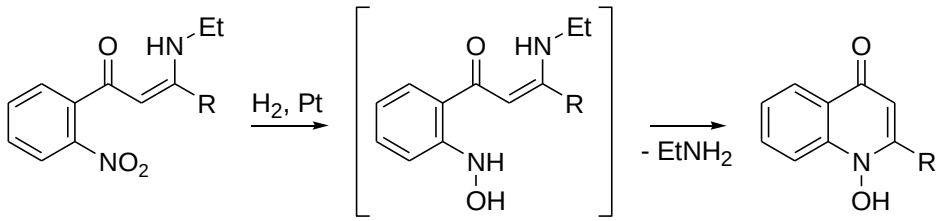
Various bacterial species of genera such as *Pseudomonas*, *Burkholderia*, *Arthrobacter*, *Stigmatella*, and *Rhodococcus* produce 2-alkyl-4-quinolone *N*-oxides with antibiotic activities [3, 4, 5]. The widely accepted

model of their antibiotic effect is based on the disruption of the respiratory chain of the competing microorganisms [6, 7]. The interesting biological profile of these compounds has motivated the development of synthetic methods for preparation of natural AQNOs and structural analogs as potential antibacterials. Until now, in the literature there have been two general approaches to the synthesis of AQNOs, both pioneered by Cornforth and James [8]. The first one includes a preparation of nitrobenzoylketones and their partial reduction with $SnCl_2$, followed by spontaneous cyclisation to AQNOs. The second approach extends the Conrad-Limpach synthesis of AQs with three additional steps (*O*-protection/*N*-oxidation/*O*-deprotection) to finally give AQNOs. Due to its inefficiency, the former approach has fallen out of favour, while the latter has been the preferred one in virtually all studies of AQNOs ever since [9, 10, 11, 12, 13]. Although there have been attempts for its optimization and refinement [10], this oxidative approach is still far from ideal and is not well suited for preparation of functionalized analogs.

In a recent publication on the synthesis of AQs, as a side result we have also described a synthesis of some carboxamide analogs of the AQNOs by partial reduction of 2-nitrobenzoylated β-enamino amides under *Pd*-catalyzed transfer hydrogenation conditions [14]. These conditions however did not provide complete control on the degree of hydrogenation and were not compatible with chlorinated substrates. Also, the structurally simpler natural AQNOs were completely out of the scope of this method, because of the poor control on the degree of hydrogenation.

Results and discussion

Considering the drawbacks of our previously published procedure, and taking into account the general interest in convenient preparative methods for AQNOs, we continued our efforts in this direction by screening a variety of conditions for chemoselective partial reduction of the nitro group in aromatic substrates. Particularly, we were interested in developing a procedure for the partial reduction of 2-nitrobenzoyl enamines **1** (Scheme 2), available through a convenient acylation/decarbamoylation of β-enamino amides [14]. The required procedure was expected not only to stop the reduction of the target substrates at the hydroxylamine level (**2**), but also to not reduce the subsequently formed products **3** (Scheme 2).



1 **2** **3**

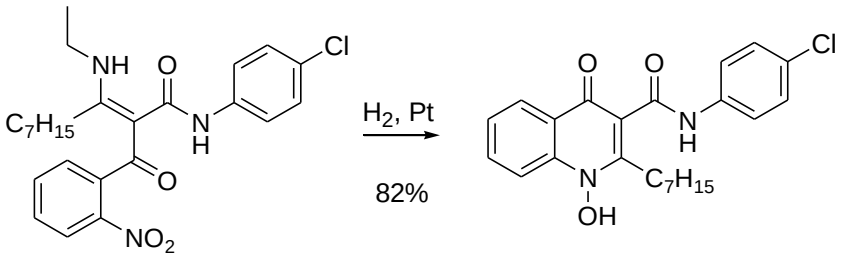
3	R	Yield %
a	*n*-C_3H_7	70
b	iBu	58
c	*n*-C_5H_{11}	70
d	*n*-C_7H_{15}	68

Scheme 2. *Synthesis of AQNOs. Reagents and conditions:* H_2 *(balloon),* 5 wt% *Pt/*Al_2O_3*, n-BuNH*$_2$*, DMSO, isopropanol, 24 h, r.t.*

After many experiments, we found that the conditions reported by Takenaka *et al.* for preparation of aromatic hydroxylamines [15] give excellent results in the context of AQNOs preparation from 2-nitrobenzoyl enamines **1**. This partial hydrogenation procedure is carried out with H_2 at atmospheric pressure and uses a combination of DMSO and *n*-butylamine to fine-tune the activity of the alumina-supported *Pt* catalyst. When the 2-nitrobenzoyl enamines **1** were subjected to these conditions, the reaction led directly to the desired *N*-oxides **3** in very good yields and purity. The intermediately formed hydroxylamines **2** (Scheme 2) cyclized spontaneously and could not be isolated. The products **3** were easily isolated after filtration of the catalyst, evaporation of the solvent under reduced pressure and a simple extractive workup, removing the residual DMSO and *n*-butylamine.

The NMR spectra of the obtained AQNO products **3** were measured at 25 °C in DMSO-d_6 and were indicative of the 4-hydroxyquinoline-*N*-oxide tautomeric form (Scheme 1, **B**), with the only exception of the poorly soluble **3b**, which had to be measured at 70 °C and appeared as the *N*-hydroxy-4-quinolone form (Scheme 1, **A**). This is in contrast to their reduced AQ analogs, which show preference for the 4(1*H*)-quinolone form in the same solvent at 25 °C [14]. The ^{1}H-NMR signals that are most indicative of the tautomeric form are those of the C3-H protons, appearing around 6 ppm for the keto tautomers and at 7.02 – 7.15 for the aromatized 4-hydroxy *N*-oxide tautomers. In the ^{13}C-NMR spectra, the most significant difference between the tautomers is observed in the chemical shifts of the C4 signals (174 – 177 ppm for a carbonyl C4, and 166 – 167 ppm for an aromatic C4-OH). Interestingly, all previously reported spectra of the known products **3c** [11] and **3d** [10, 11] have been measured in deuterated methanol and indicate a preference for the *N*-hydroxy-4-quinolone form in this solvent, with the C4-carbonyl ^{13}C signal visible only in HMBC.

After the successful preparation of the *N*-oxides **3**, we tried the same hydrogenation conditions with the chlorinated carboxamide substrate **4**. In this case the procedure also worked excellently and gave the expected product **5** in 82% yield, without any concomitant reduction at the C–Cl bond (Scheme 3).



4 **5**

Scheme 3. *Synthesis of a halogenated carboxamide analog (**5**) of the P. aeruginosa metabolite HQNO.*

In conclusion, we have described a chemoselective and operationally simple synthesis of 4-hydroxyquinoline *N*-oxides from 2-nitrobenzoyl enamines. One of the obtained products (**3d** or HQNO) is a known bacterial toxin, produced by *P. aeruginosa*. The synthetic procedure is also compatible with chlorinated substrates and extends the range of accessible analogs for biological studies.

Experimental

The preparation of the starting compounds **1** and **4** is described in detail elsewhere [14]. Unless otherwise noted, all other reagents and solvents were purchased from Sigma-Aldrich, Darmstadt, Germany, and were used as supplied. NMR spectra were run on Bruker NEO 400 (400/100 MHz ^{1}H/^{13}C) spectrometer. Chemical shifts (δ, ppm) are downfield from TMS. TLC was done on aluminium-backed Silica gel 60 sheets (Merck) with $KMnO_4$ staining. Melting point measurements were done in capillary tubes on KRÜSS M5000 automatic mp meter and are not corrected.

*Synthesis of compounds **3** and **5**, general procedure:* The corresponding starting compound **1** (0.5 mmol) or **4** (0.236 g, 0.5 mmol) was dissolved in isopropanol (IPA) (2 mL). Then DMSO (0.013 g, 0.012 mL), *n*-butylamine (0.037 g, 0.050 mL) and 5 wt% *Pt/Al_2O_3* (0.010 g) were added to the solution. The air in the reaction vessel was evacuated and replaced with H_2 with the help of a three-way stopper. The H_2 atmosphere was kept with a balloon for the next 24 h, while the reaction mixture was magnetically stirred at 25 °C. Then the catalyst was filtered off through a pad of celite on a sintered glass funnel, with thorough rinsing with IPA. The IPA was

removed from the filtrate on a rotary evaporator under reduced pressure. To remove any residual *n*-butylamine and DMSO, dilute aqueous *HCl* was added to the residue and the product was extracted in CH_2Cl_2 (2 × 30 mL). The combined organic layers were dried with anhydrous sodium sulfate, the solvent was removed under reduced pressure and the residue was purified by column chromatography on a short silica gel plug, using diethyl ether as the eluent (increasing polarity to Et_2O:MeOH 10:1). Once crystal seeds are available, the products could be isolated by simple trituration with diethyl ether and seeding to facilitate the crystallization of the product.

*2-Propyl-4-hydroxyquinoline N-oxide (**3a**):* Yield: 72 mg (70%), white solid, m.p. 154–155 °C; ^{1}H-NMR (DMSO-d_6, δ ppm, *J* Hz): 1.01 (t, 3H, *J* = 7.4), 1.79 (sext, 2H, *J* = 7.4), 3.07 (t, 2H, *J* = 7.4), 7.02 (s, 1H), 7.75 (m, 1H), 8.05 (m, 1H), 8.24 (m, 1H), 8.29 (m, 1H); ^{13}C-NMR (DMSO-d_6, δ ppm): 14.1, 21.0, 33.5, 105.6, 117.2, 121.5, 124.3, 127.5, 134.7, 140.1, 158.2, 167.6; Anal. Calcd for $C_{12}H_{13}NO_2$: C, 70.92; H, 6.45; N, 6.89. Found: C, 70.98; H, 6.51, N, 6.78.

*1-Hydroxy-2-isobutylquinolin-4(1H)-one (**3b**):* Yield: 63 mg (58%), white solid, m.p. 230–231 °C (dec.); ^{1}H-NMR (70 °C, DMSO-d_6, δ ppm, *J* Hz): 0.97 (d, 6H, *J* = 6.7), 2.16 (m, 1H), 2.66 (d, 2H, *J* = 7.2), 5.97 (s, 1H), 7.38 (m, 1H), 7.73 (m, 1H), 7.87 (m, 1H), 8.12 (m, 1H); ^{13}C-NMR (70 °C, DMSO-d6, δ ppm): 22.6, 27.2, 40.3, 108.3, 115.5, 123.9, 125.3, 125.4, 132.2, 141.0, 152.7; Anal. Calcd for $C_{13}H_{15}NO_2$: C, 71.87; H, 6.96; N, 6.45. Found: C, 72.02; H, 7.03, N, 6.41.

*2-Pentyl-4-hydroxyquinoline N-oxide (**3c**):* Yield: 81 mg (70%), white solid, m.p. 139–140 °C; ^{1}H-NMR (DMSO-d_6, δ ppm, *J* Hz): 0.89 (t, 3H, *J* = 7.0), 1.36 (m, 4H), 1.77 (m, 2H), 3.12 (t, 2H, *J* = 7.8), 7.16 (s, 1H), 7.80 (m, 1H), 8.09 (m, 1H), 8.30 (m, 2H), 11.70 (br s, 1H); ^{13}C-NMR (DMSO-d_6, δ ppm): 14.2, 22.2, 27.2, 31.2, 31.7, 105.4, 117.4, 121.1, 124.2, 128.0, 135.0, 140.0, 158.9, 166.8; Anal. Calcd for $C_{14}H_{17}NO_2$: C, 72.70; H, 7.41; N, 6.06. Found: C, 72.57; H, 7.48, N, 5.92.

*2-Heptyl-4-hydroxyquinoline N-oxide (**3d**):* Yield: 88 mg (68%), white solid, m.p. 159–160 °C; ^{1}H-NMR (DMSO-d_6, δ ppm, *J* Hz): 0.85 (t, 3H, *J* = 6.8), 1.21 – 1.42 (m, 8H), 1.75 (m, 2H), 3.11 (t, 2H, *J* = 7.8), 7.15 (s, 1H), 7.79 (m, 1H), 8.09 (m, 1H), 8.30 (m, 2H), 11.65 (br s, 1H); ^{13}C-NMR (DMSO-d_6, δ ppm): 14.4, 22.5, 27.5, 28.8, 29.1, 31.6, 31.8, 105.4, 117.4, 121.1, 124.2, 127.9, 135.0, 140.0, 158.9, 166.9; Anal. Calcd for $C_{16}H_{21}NO_2$: C, 74.10; H, 8.16; N, 5.40. Found: C, 73.97; H, 8.20, N, 5.35.

*1-Hydroxy-2-heptylquinolin-4(1H)-one-3-carboxylic acid (4-chlorophenyl)-amide (**5**):* Yield: 170 mg (82%), white solid, m.p. 207–208 °C; ^{1}H-NMR (DMSO-d_6, δ ppm, *J* Hz): 0.81 (t, 3H, *J* = 6.9), 1.17 – 1.32 (m, 6H), 1.38 (m, 2H), 1.75 (m, 2H), 3.08 (m, 2H), 7.38 (m, 2H), 7.48 (m, 1H), 7.75 (m, 2H), 7.83 (m, 1H), 7.93 (m, 1H), 8.23 (m, 1H), 11.39 (s, 1H), 12.12 (s, 1H); ^{13}C-NMR (DMSO-d_6, δ ppm): 14.4, 22.5, 28.4, 28.7, 29.2, 29.5, 31.5, 114.9, 115.7, 121.4, 125.0, 125.5, 125.9, 127.1, 129.1, 133.4, 138.8, 139.9, 155.7, 164.7, 173.5; Anal. Calcd for $C_{23}H_{25}ClN_2O_3$: C, 66.90; H, 6.10; N, 6.78. Found: C, 66.80; H, 6.17, N, 6.72.

Acknowledgement

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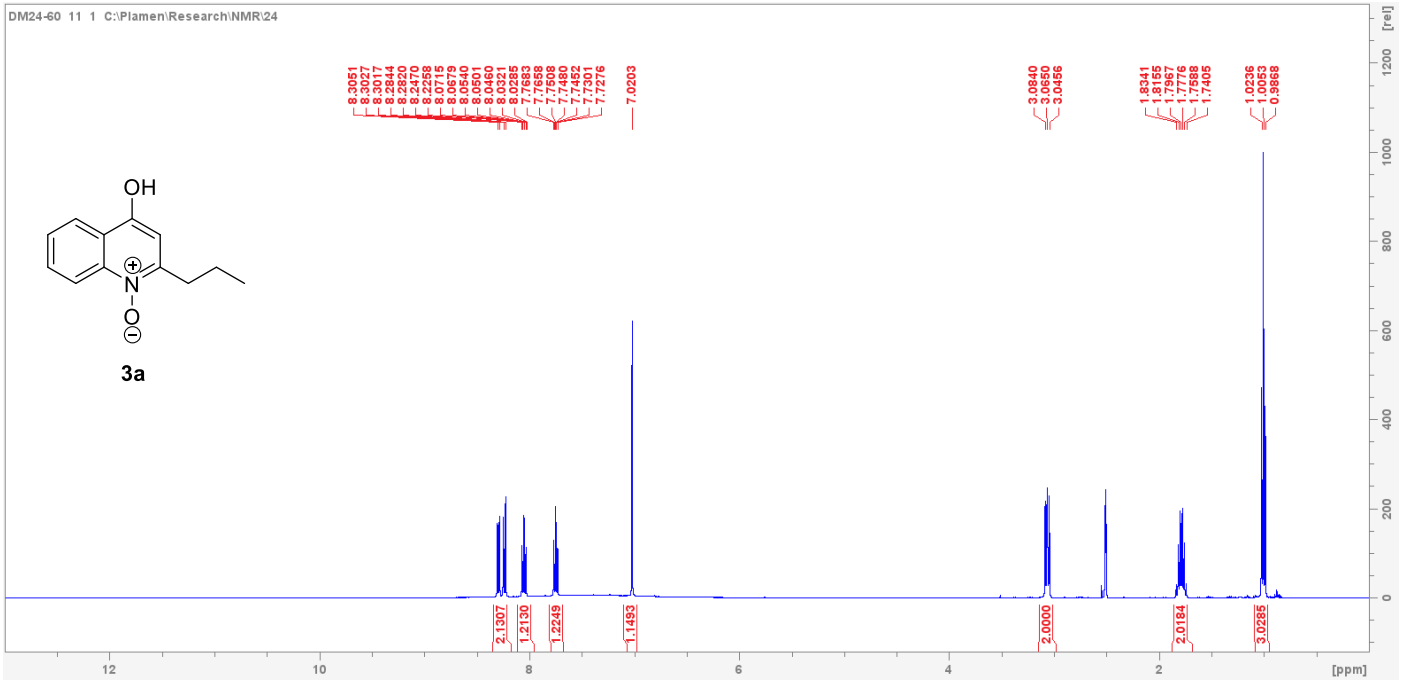
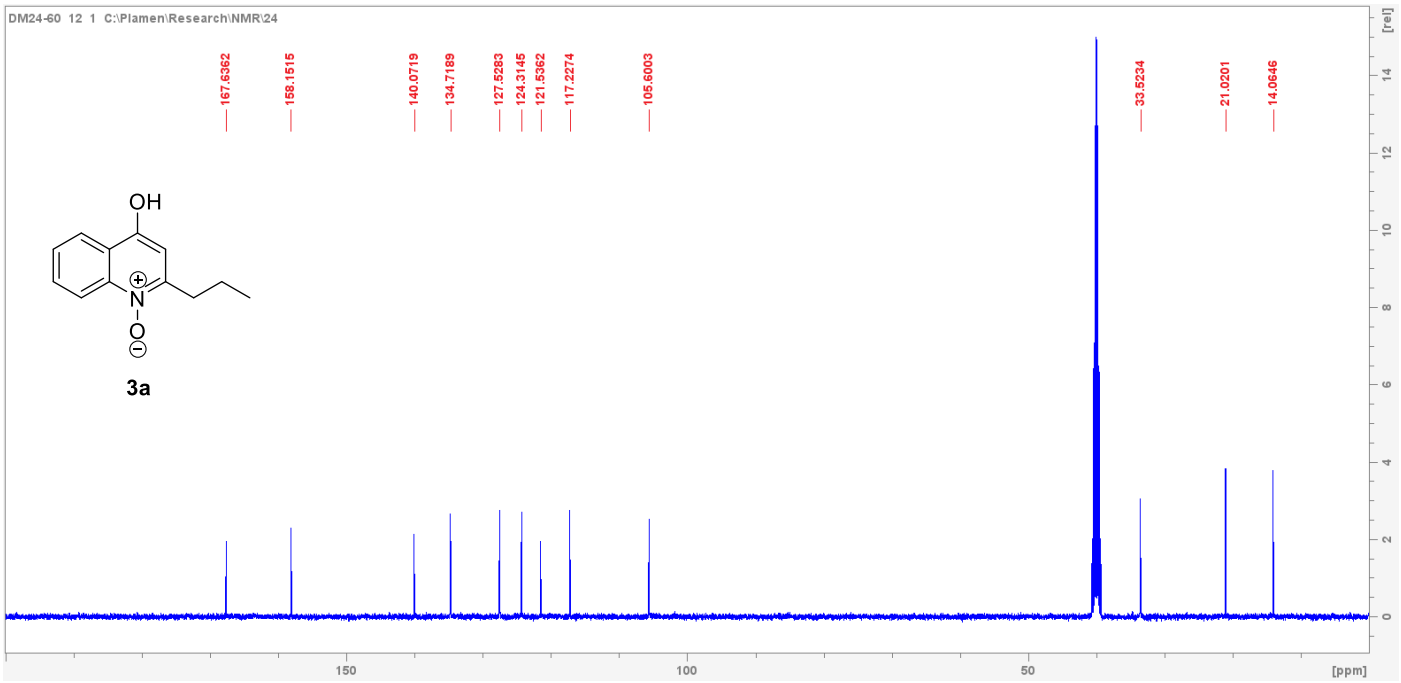
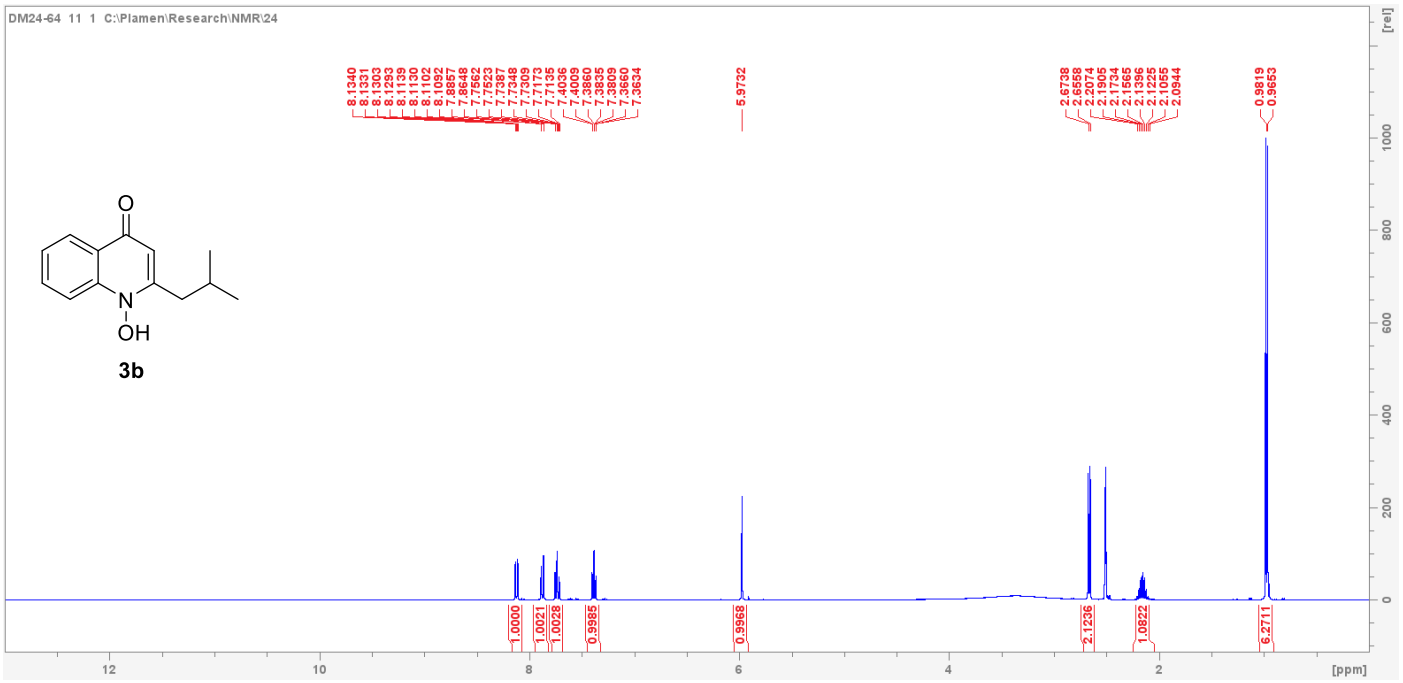
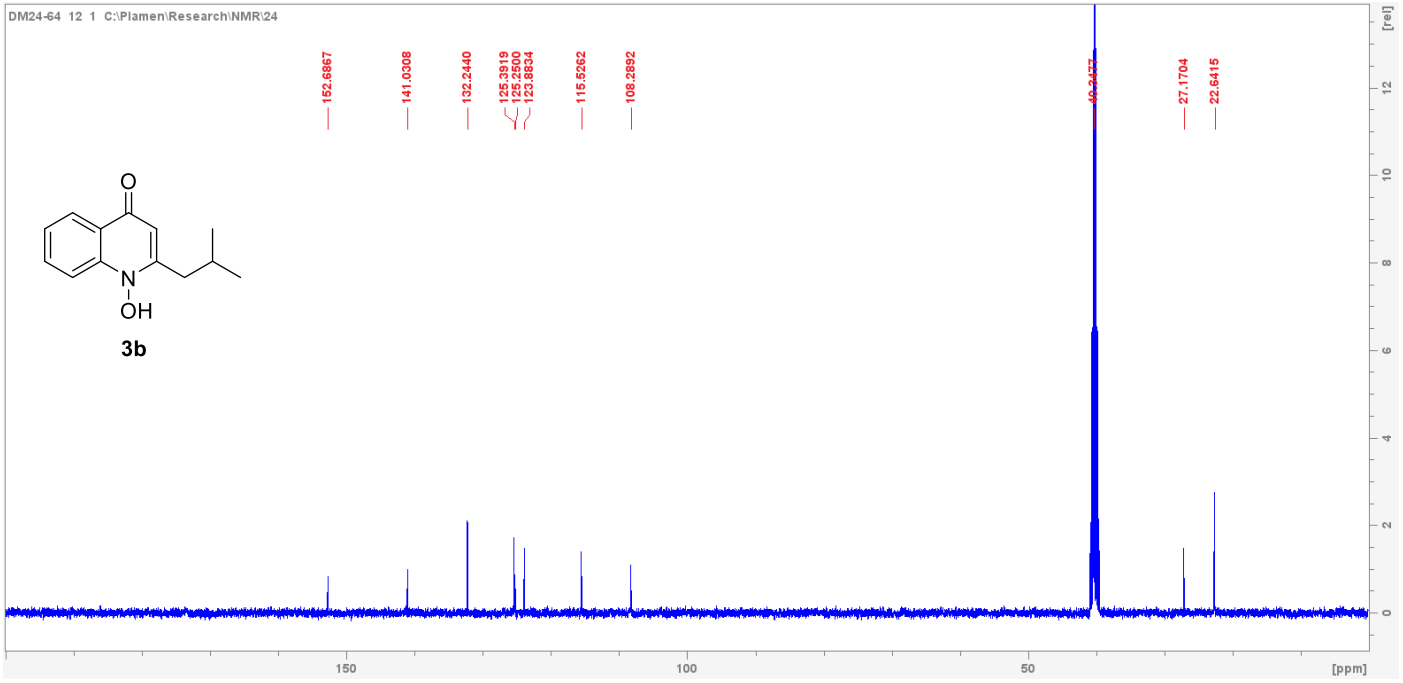
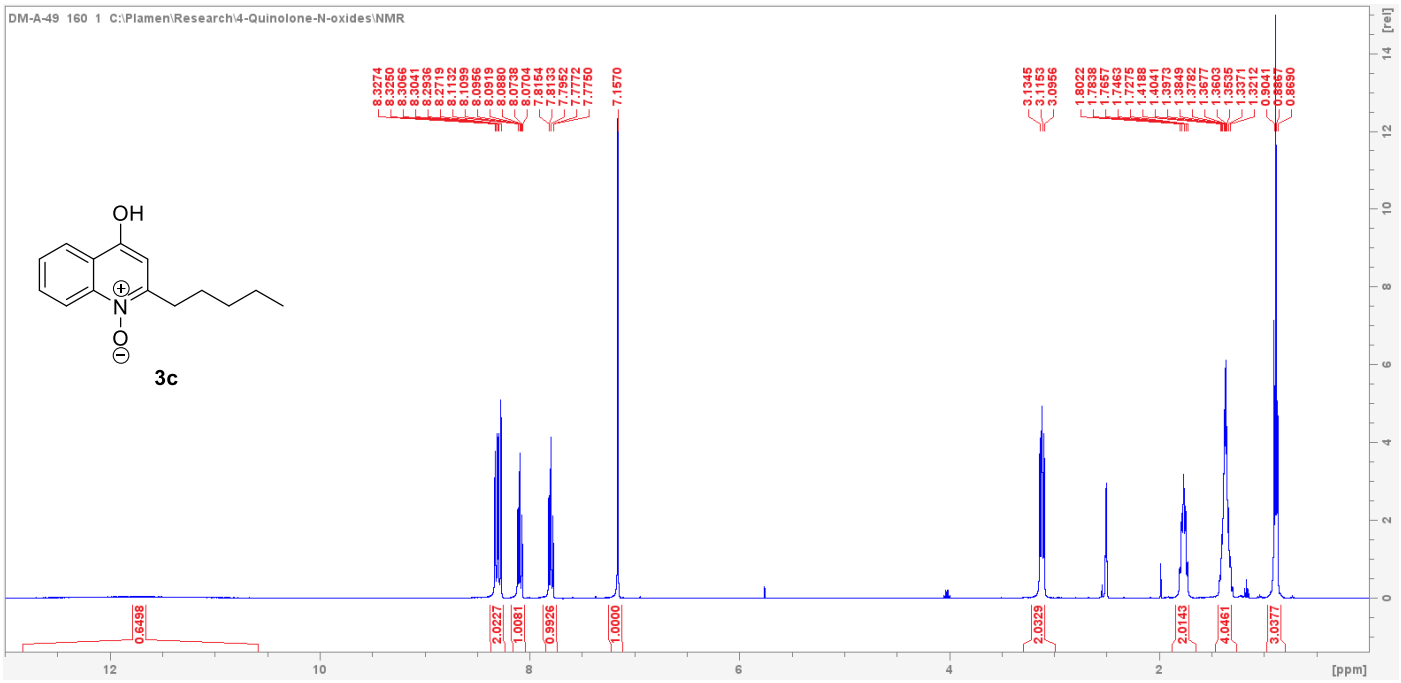
^{1}H and ^{13}C NMR spectra

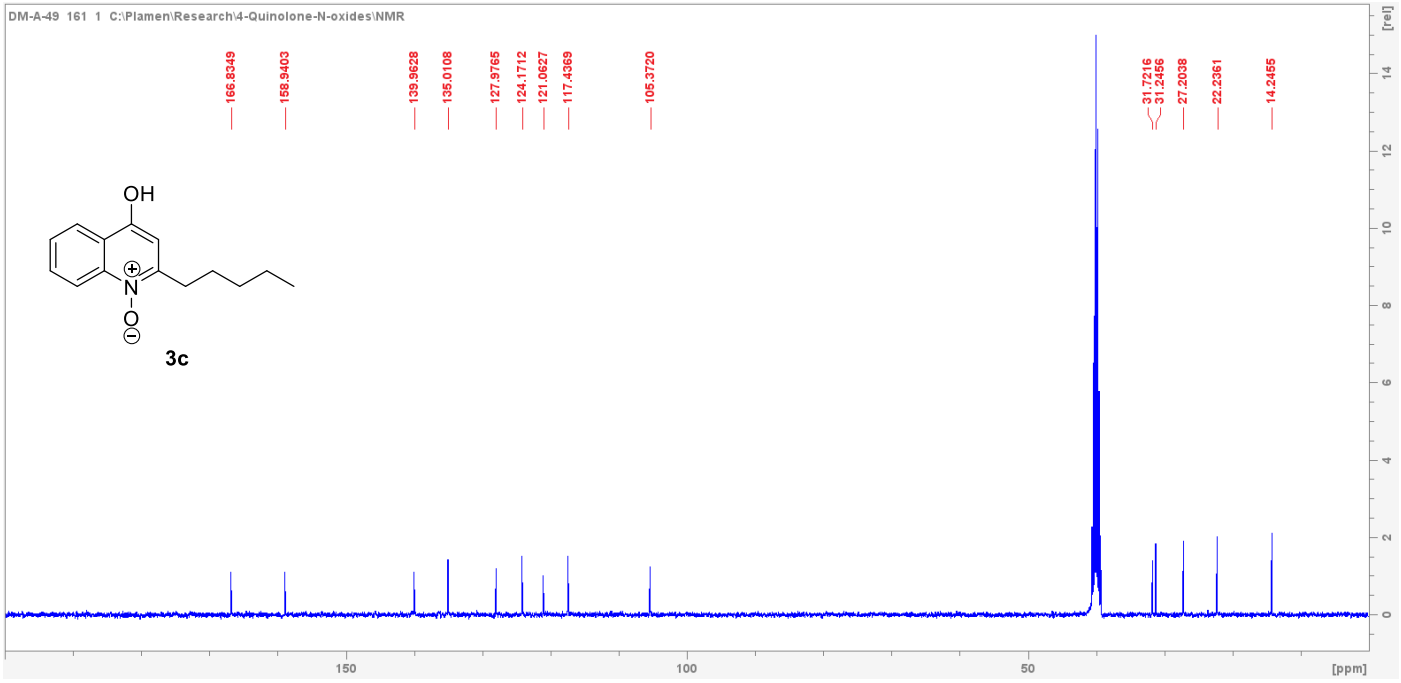


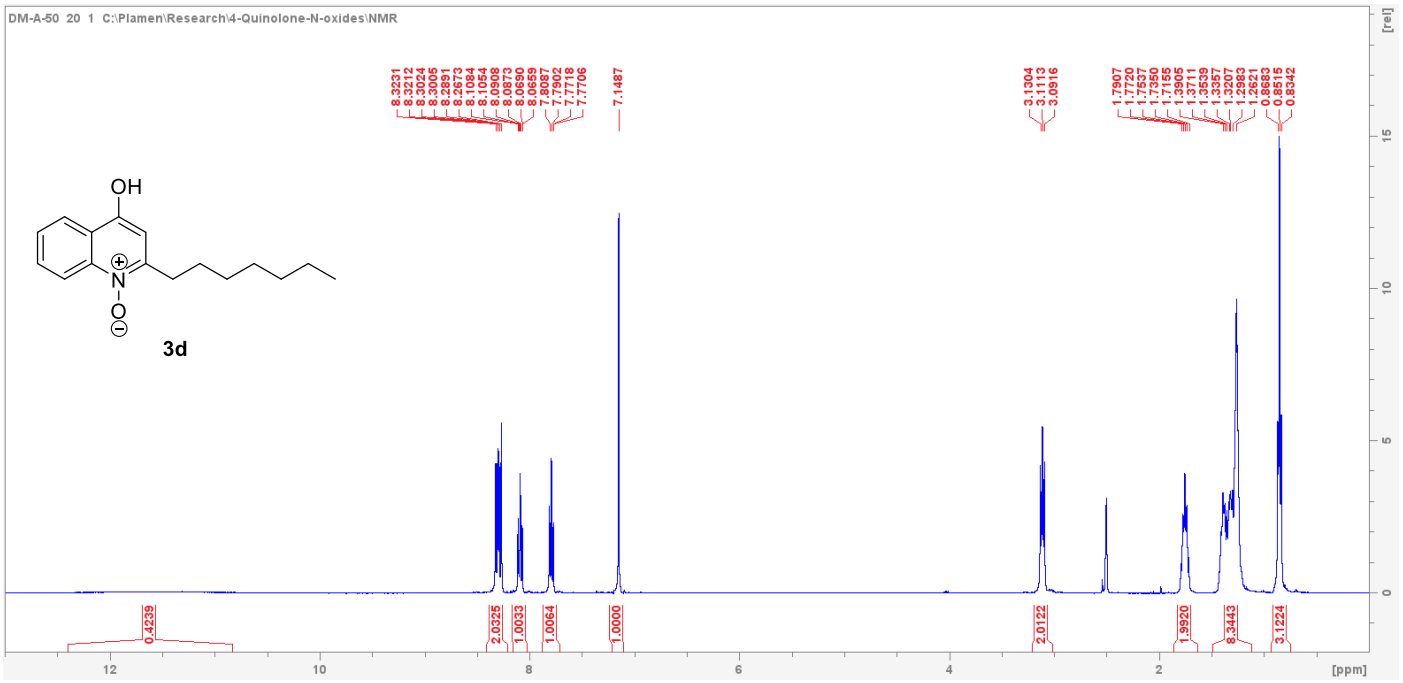


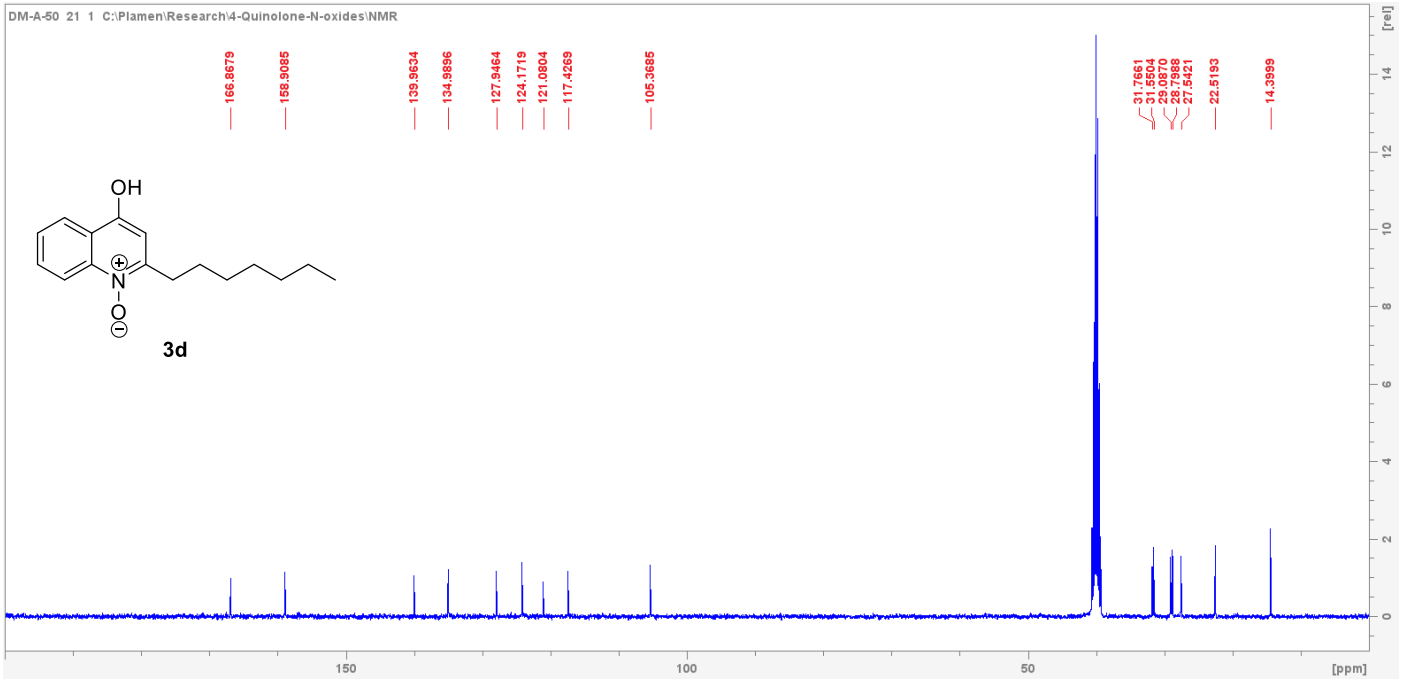


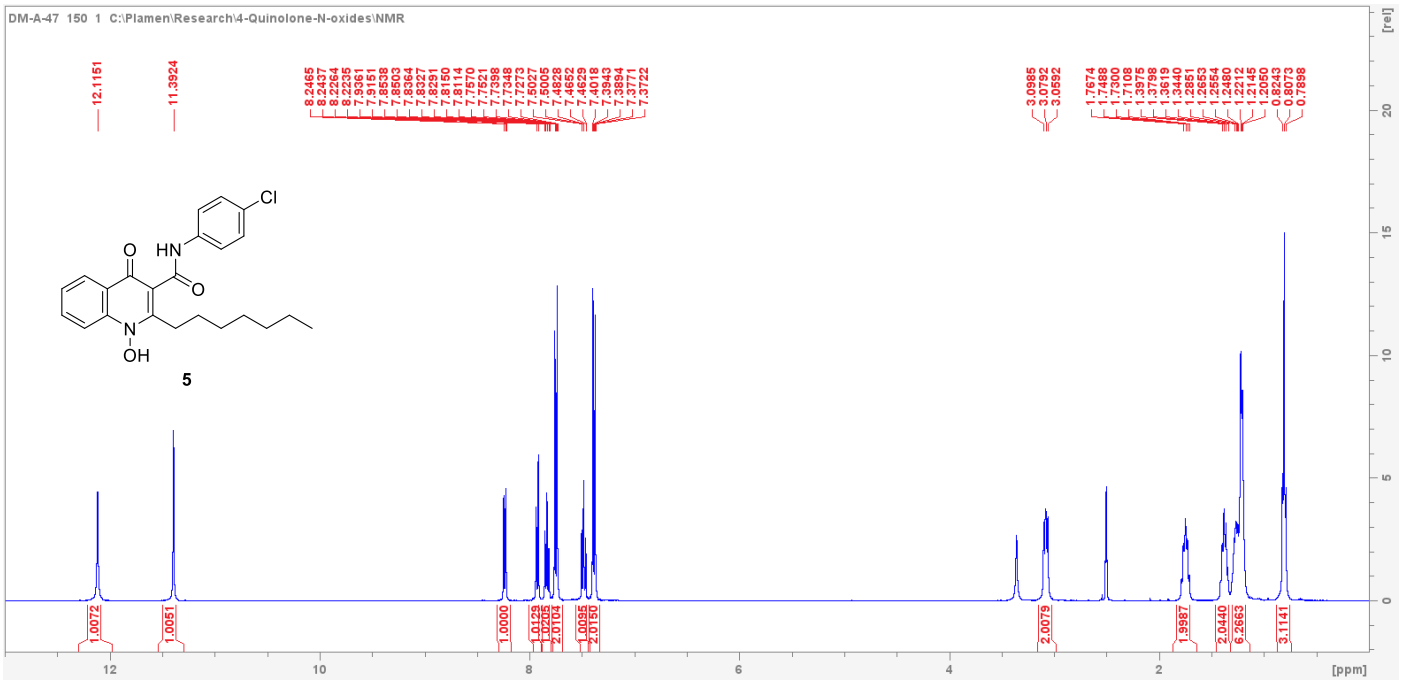


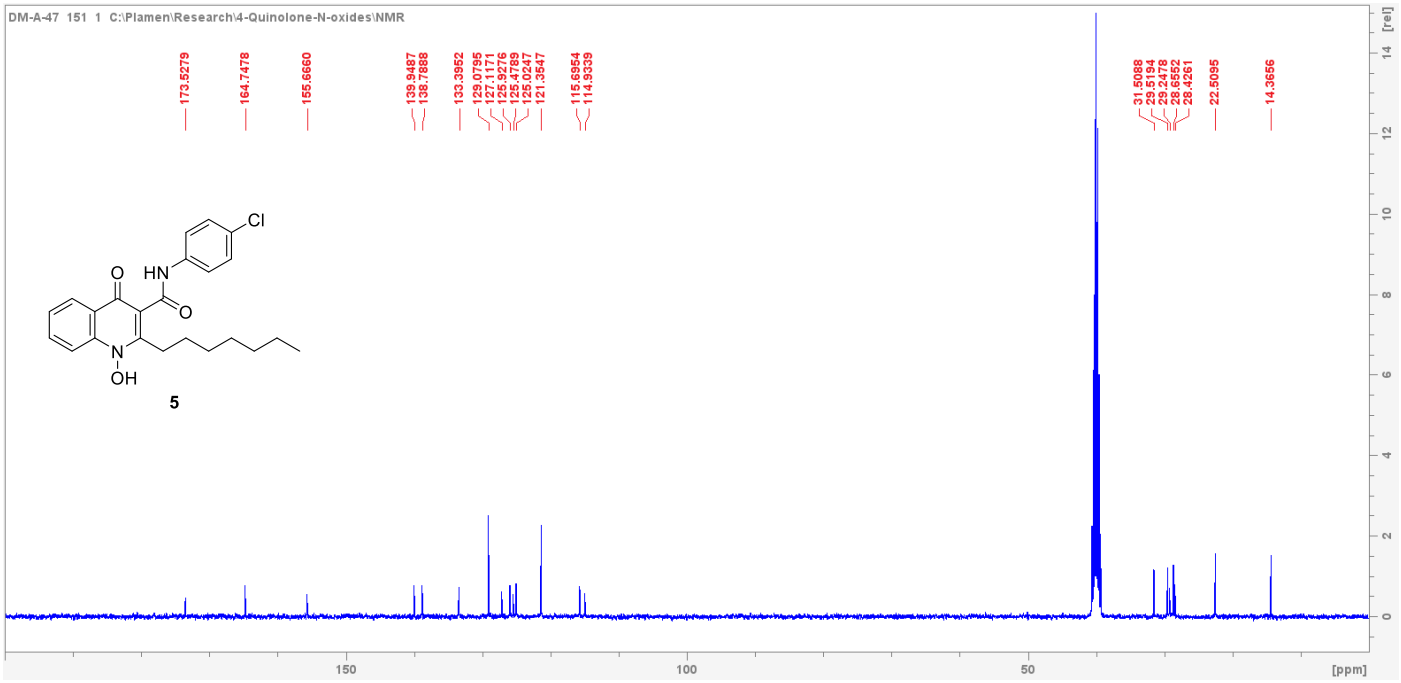






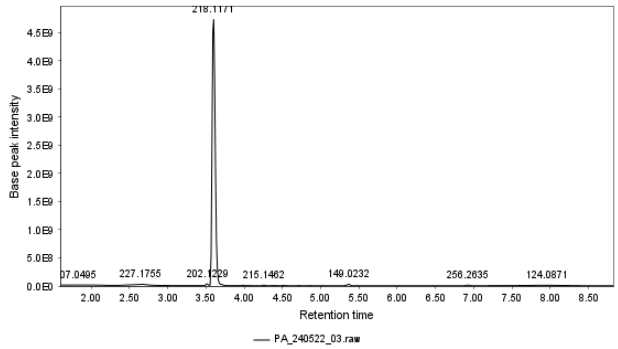


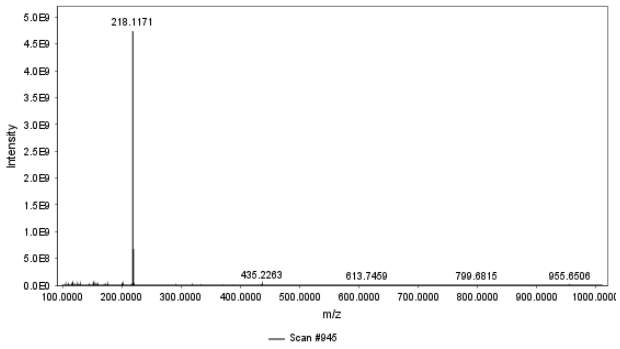




HPLC-MS (ESI+)

*1-Hydroxy-2-isobutylquinolin-4(1H)-one (**3b**):* calcd. for $C_{13}H_{16}NO_2^+$ $[M+H]^+$ 218.1176, found 218.1171





*1-Hydroxy-2-heptylquinolin-4(1H)-one-3-carboxylic acid (4-chlorophenyl)-amide (**5**):*
calcd. for $C_{23}H_{26}ClN_2O_3^+$ $[M+H]^+$ 413.1626, found 413.1617